

# **APPENDIX 1**

## **Public and Peer Review Panel Comments**

**Appendix 1-3**  
**Authors Responses to Comments**

**Appendix 1-3g**  
**Author's Response to Comments on Chapter 7**

## **Chapter 7: The Everglades Mercury Problem**

### **Response to Exponent on Chapter 7 of ECR 2000 from March 31, 2000**

Prepared by: Larry Fink and Darren Rumbold

To assist in the preparation of the Everglades Consolidated Report (ECR) 2001, the public was provided with an opportunity to provide further comment on last year's report by March 31, 2000, and to submit substantive reports that the District should consider in the preparation of this year's report by July 1, 2000. This section is devoted to comments and responses on Chapter 7: The Everglades Mercury Problem and the associated appendices. Chapter 7 summarizes the Everglades mercury problem, the multi-agency program for understanding and solving it, and the status and key findings of the individual monitoring, research, modeling, and assessment projects that have been completed, are ongoing, or planned to implement the program and achieve its goals. This section first summarizes statutory requirements for pollutants other than phosphorus, including mercury, then summarizes District responses to comments received on last year's Chapter 7 by March 31, 2000.

#### **Statutory Requirements for Pollutants other than Phosphorus**

For pollutants other than phosphorus, including total mercury (THg), the EFA requires:

(4)(d) 1. By January 1996, the department and the district shall review and evaluate available water quality data and identify any additional information necessary to adequately describe water quality .... in the Everglades Protection Area and tributary waters.

(4)(d) 1 ... the district shall monitor all discharges into the Everglades Protection Area for purposes of determining compliance with state water quality standards.

(4)(d) 2. The research and monitoring program shall ... evaluate water quality standards ...

(4)(d) 3. The research and monitoring program shall include research seeking to optimize the design and operation of the STAs ....

(4)(d) 5. Beginning January 1, 2000, the district and the department shall annually issue a peer-reviewed report (on) the research and monitoring program that summarizes all data and findings.

(4)(e) 1. The department and the district shall ... complete (no later than December 31, 2001) any additional research necessary to: ... b. Evaluate existing water quality standards ....

(4)(e) 3. The department shall use the best available information to define relationships between waters discharged to, and the resulting water quality in, the Everglades Protection Area..

(4)(e) 3. ... The department or the district shall use these relationships to establish discharge limits in permits ... to prevent an imbalance in the natural populations of aquatic flora or fauna in the Everglades Protection Area and provide a net improvement in the areas already impacted. ...

(4)(e) 4. The department's evaluation of any other water quality standards must include the department's antidegradation standards and EAA canal classifications.

(4)(f) 1. The district, in cooperation with the department, shall develop and implement a water quality monitoring program to evaluate the effectiveness of the BMPs in achieving and maintaining compliance with state water quality standards and restoring ... beneficial uses. ...

(4)(f) 2. ... implement a comprehensive program of research, testing, and implementation of BMPs that will address all water quality standards ...

(4)(f) 4. As of December 31, 2006, ... no permittee's discharge shall cause or contribute to any violation of water quality standards in the Everglades Protection Area.

9 (h) Discharges shall be allowed, provided the STAs are operated in accordance with this section, if, after a stabilization period:

The STAs achieve the design objectives of the Everglades Construction Project for phosphorus;

For water quality parameters other than phosphorus, the quality of water discharged from the STAs is of equal or better quality than inflows; and

Discharges from STAs do not pose a serious danger to the public health, safety, or welfare.

## **Response to Exponent Comments from March 31, 2000 on Chapter 7**

This is in response to comments received from Gary Bigham of Exponent on March 31, 2000, on deficiencies in last year's Chapter 7: The Everglades Mercury Problem in the Everglades Consolidated Report (ECR 2000) that need to be addressed in the preparation of this year's Chapter 7 (ECR 2001). Exponent is under contract to the Sugar Cane Growers Cooperative of Florida.

### **General Comments**

Interwoven with the specific comments are several general concerns that Exponent has asked to be corrected in this year's Chapter 7: (1) the use of deductive reasoning to arrive at conclusions rather than hypotheses, (2) subjectivity and advocacy in evaluating post-ECP Hg impacts, and (3) the excessive use of personal communications. Below we address these general concerns, and then each specific concern is addressed in the order received.

#### **• The Use of Deductive Logic**

Comment: "In general, the (District's) presentation is a holistic exercise in deductive logic," "...in addressing matters such as mercury fate and transport."

Response: Most (> 80%) of last year's Chapter 7 was devoted to:

- a summary of mercury research strategies, programs, and projects;
- a summary of the analysis of empirical data gathered to quantify STA mercury removal efficiencies
- a summary of the analysis of empirical data to quantify mercury pathways, concentrations, and risk status and trends; and
- evaluation of the adequacy of the existing numerical Class III Water Quality Standard for the protection of human health and terrestrial wildlife.

To predict post-ECP mercury impacts, the District assumed that the pre-ECP mercury conditions at unimpacted WCA-2A-U3 are representative of and a surrogate for post-ECP restored conditions at presently eutrophic sites like WCA-2A-F1. This is the so-called reference site approach. Once that assumption is accepted, no further use of deductive logic is required regarding mercury sources, transport, and fate to predict post-ECP mercury impacts.

- **The District limited its use of deductive logic to the:**
- invalidation of Commenter's hypothesis that a decrease in TP-mediated biodilution was the cause of the observed increase in mosquitofish THg with downstream distance along the "F" research transect in WCA-2A.
- invalidation the univariate empirical model calibrated with "F" transect TP and mosquitofish THg data for predicting post-ECP mercury impacts.
- development of a mechanistic explanation for the influences of elements and compounds other than TP on MeHg production or bioaccumulation along the WCA-2A nutrient gradient to evaluate the margin of safety in the reference site-based estimate of post-ECP mercury risks.

These analyses and predictions were carried out on pages 7-41 to 7-65 of last year's Chapter 7.

Comment: "Although this is appropriate for generation of scientific hypotheses, it is contrary to the scientific method to use it to develop conclusions."

Response: The extrapolation or prediction of post-operational mercury impacts from ECP Phase 1 and 2 technologies must be based on and not be inconsistent with accepted scientific hypotheses and valid published and unpublished data. However, the extrapolation or prediction itself is not the product of empirical science per se, because the future cannot be measured. Nevertheless, such extrapolations or predictions must still be carried out in a logical, systematic, and self-consistent way without contradicting scientific laws, well-established hypotheses, and extant relevant data. In the scientific and engineering arena, this is usually accomplished qualitatively with conceptual models and quantitatively with empirical or mechanistic mathematical models. The results generated by such models are the product of deductive reasoning and the conclusions drawn from the model results are used to guide restoration decision-making. This is the case for every other element of ECP and CERP, and the mercury restoration effort should not be treated differently.

Where the District engaged in qualitative or quantitative predictions using conceptual or mathematical models, the District made its assumptions, logic, and data explicit, cited supporting scientific evidence, and discussed such logical explanations in terms of what may or could occur, not in terms of what will occur. The District also cited contrary hypotheses and conflicting data to ensure balance in the discussion. The District characterized the confidence in its predictions using adjectives such as “highly unlikely,” “unlikely,” “likely,” and “highly likely” to guide restoration decision-makers. Last year’s peer review panel reviewed the District’s assessment of post-ECP mercury impacts and found no serious deficiencies in methods, data capture, reduction, analysis, or presentation, or findings, conclusions, or recommendations. Nevertheless, to ensure that there is no ambiguity in this regard, when formulating an explanation for empirical observation, the District will use the following clarifying phrase: “It can be hypothesized with a limited, moderate, or high degree of confidence that ....”, and when predicting an effect from a cause that has not yet occurred, “It can be speculated with a limited, moderate, or high degree of confidence that ....”

However, contrary to Commenter’s statement, the use of deductive logic to falsify or invalidate hypotheses is not inconsistent with the scientific method. For example, the District used deductive logic to invalidate Commenter’s “biodilution hypothesis” (Pages 7-54 to 7-56, ECR 2000) as an explanation for the observed exponential increase in mosquitofish THg with downstream distance along the “F” research transect in the nutrient-impacted area in WCA-2A (Exponent, 1998; 1999; 2000a). To do this, the District obtained valid empirical data from the “F” transect that were inherently inconsistent with this hypothesis. The discussion of the hypothesis and its invalidation are taken up later in the responses to the specific comments about the inverse relationship with phosphorus and the THg flux model.

Comment: “... 2001 report (should) take a more objective and less advocative position by presenting the products of deduction as hypotheses, not conclusions.”

Response: The District does not agree that it was either subjective or advocative in the use of deductive reasoning to falsify Exponent’s biodilution hypothesis or to invalidate the use of its univariate empirical model for predicting post-ECP MeHg risks to fish-eating animals. Having evaluated all of the evidence before it, the peer review panel concluded that the District’s conceptual model based on the predominant influence of sulfur, not phosphorus, on the mercury cycle was the more robust, accurate, and scientifically defensible. However, whether the District’s mechanistic explanations for sulfur’s influences are ultimately proven to be incorrect is of no consequence to the validity of the District’s predictions of post-ECP risks to fish-eating wildlife. This is because the District relied solely on a reference site approach using unimpacted WCA-2A-U3 as a surrogate for restored eutrophic sites like WCA-2A-F1 to predict its post-ECP ecological risks. No further deductive reasoning or mechanistic explanations were required. Mechanistic hypotheses and models were developed and applied only to evaluate the margin of safety in the reference site predictions.

- **THE USE OF PERSONAL COMMUNICATIONS**

Comment: “A general concern with the 2000 report that Exponent would like to be remedied in the 2001 report is the dependence of the District’s arguments on personal communications. In the section on inverse relationships alone, this occurred eighteen times. Such hearsay cannot be considered evidence because it has not been subjected to validation or peer review. We, therefore, request that any time the authors must rely on unpublished data that they make those data available for review either within the chapter or in an appendix.”

Response: Quality-assured data collected by recognized experts following standard methods and procedures should not be withheld from restoration decision-makers solely because they have not yet been published in the open, peer-reviewed scientific or engineering literature. Awaiting such publication could delay access to crucial results by several years, which is often outside the window of opportunity to influence timely restoration management decision-making. As is the case with unpublished data, the information conveyed in personal communications included in the Everglades Consolidated Report are reviewed by the peer review panel to ensure their scientific accuracy. We anticipate that the results and conclusions conveyed in personal communications and unpublished agency reports will ultimately be published in credible, peer-reviewed scientific or engineering journals over the next several years, reducing the need to make use of unpublished data and personal communications over time. In the interim, we will continue to make use of personal communications and unpublished data from recognized scientific experts and agency officials to the extent deemed appropriate and necessary by the District to meet its general and specific Everglades restoration and protection mandates in state and federal statutes and permits in a timely fashion.

All of the personal communications in last year's Chapter 7 are from agency officials or recognized experts in the field who will testify to the findings or conclusions transmitted as personal communications if this becomes necessary. The administrative or judicial rules of evidence and applicable precedents in allowing and evaluating the mainstream scientific credibility of expert testimony will then apply. Such expert testimony does not constitute hearsay, and the rules regarding the admissibility of hearsay evidence do not apply.

Finally, Commenter has miscounted the number of personal communications and references to unpublished data in the section on the inverse relationship. There were six such references, not eighteen.

## **SPECIFIC COMMENTS**

### **• WATER COLUMN PHOSPHORUS CONCENTRATIONS**

Comment: "One example is the suppositions on pages 7-37 to 7-38 concerning the role of dissolved oxygen concentrations in mercury methylation."

Response: MeHg production occurs under anaerobic conditions and, thus, cannot be divorced from the dissolved oxygen (DO) regime in the surficial sediment, which, in turn, cannot be divorced from the DO regime in the overlying water. Commenter's issues regarding the role of DO are not new, have been adequately addressed on page 7-38 of Chapter 7 and on pages A1-3-74 and 75 of Appendix 3 of last year's Everglades Consolidated Report (2000), and the District responses do not need to be repeated here.

### **• INVERSE RELATIONSHIP WITH PHOSPHORUS**

Comment: "The District has laudably begun to take a mechanistic approach in looking at the potential relation between eutrophication and methylmercury bioaccumulation."

Response: This is not a new position for the District. (See for example District responses to challenges to the ENR Project NPDES permit filed by the Sugar Cane Growers Cooperative of Florida in the winter of 1994.)

Comment: “Exponent would have liked an opportunity to comment on this material during the review process, but it was not presented in the review draft.”

Response: This material was added to the September 1999 review draft of Chapter 7 at Commenter’s request to explain the observed influences of sulfur, carbon, calcium, and dissolved oxygen on MeHg production or bioaccumulation. This material was available for comment following distribution of the revised draft at the District’s November 3, 1999, public workshop. Commenter did have an opportunity to comment, albeit for a foreshortened period. The comment period ending March 31, 2000, was added to ensure that just such concerns were addressed prior to the preparation of Chapter 7 of the ECR 2001.

Comment: “In the 2000 report, the District deduced (based on no testable evidence) that the acknowledged, demonstrable inverse relationship between phosphorus concentrations in water and methylmercury bioaccumulation has no causal basis.”

Response: As propounded by Hakanson (1980) and applied by PTI (1994) to the WCA-2A “F” transect, in a P-limited aquatic ecosystem, a decrease in biodilution caused by a decrease in water column TP must manifest itself as an increase in the concentration of THg in plant biomass, all other things being equal. This is an observable phenomenon and constitutes testable evidence. While the District’s scoping calculations are based on limited data on THg in periphyton (Cleckner et al., 1998b) and emergent macrophytes (USGS-Madison, unpublished data), a single valid observation that is contrary to the stated hypothesis invalidates or falsifies it. The data and calculations presented in Table 7-1 on page 7-55 of ECR (2000) are inherently inconsistent with Commenter’s hypothesis. Therefore, water column TP-mediated biodilution cannot be the cause of the observed five-fold decrease in the concentration of THg in the periphyton community and the concomitant observed 11-fold increase in the average THg concentration in the mosquitofish community between F1 and U3. The District has concluded that this hypothesis fails because light, not water column TP, is limiting for periphyton in the eutrophic portions of the “F” transect due to shading by the dense canopies of emergent macrophytes (Grimshaw et al., 1997). However, the invalidation of the general hypothesis when applied to this specific case does not necessarily invalidate the general hypothesis when applied appropriately to other cases for which the physical, chemical, or biological particulars are different.

Comment: “The District bases this position on three suppositions. First, it is assumed that macrophyte productivity in eutrophic areas is nearly twice that in the oligotrophic portions of WCA-2A as the results of increased phosphorus inputs.”

Response: To clarify the sources of the data, macrophyte coverages at F1 and U3 were obtained from McCormick et al. (1998), macrophyte densities were measured at WCA-2A-F1 and WCA-2A-U3 by Miao and Sklar (1998), and productivity of cattail at WCA-2A-F1 and sawgrass at WCA-2A-F4 come from (Davis, 1989). At that time, F4 was an unimpacted site. Since then, the front of excess TP in water and sediment has overtaken F4, such that F4 is now a transition site and U3 is now considered the only truly unimpacted site along the “F” transect (McCormick et al., 1999; 2000). No new measurements of macrophyte productivities have been made by District staff at either F1 or U3 (S. Miao, pers. comm., 2000), so the results of the earlier Davis study at F1 and F4 were used to approximate the productivities at F1 and U3 contemporary with the other measurements used in these calculations. All of the data used by the District to calculate plant biomass turnover at F1 and U3 are available in the peer reviewed scientific literature.

Comment: “Second, although macrophytes clearly demonstrate increased productivity and reduced mercury concentrations along the phosphorus gradient, their capacity to store and turn over mercury is assumed to be insignificant with regard to total mercury fluxes.”

Response: The magnitudes of the mercury fluxes in macrophytes and periphyton were estimated from scoping-level calculations combining published data on coverages, densities, and productivities (see above) with unpublished data on THg in cattail and sawgrass stems and leaves (D. Krabbenhoft, USGS, unpublished data, 1999). Therefore, the conclusion that the THg flux mediated by macrophyte biomass turnover is a minor component of the overall plant-mediated flux of THg at F1 and U3 is derived from the described calculations.

Comment: “Third, it is assumed that the higher productivity of the periphyton in the nutrient-poor regions of the Everglades (and lower productivity in the eutrophic regions due to macrophyte shading) and associated high mercury concentrations are solely responsible for higher mercury concentrations in the oligotrophic food chain.”

Response: In developing and applying the conceptual model, the District did not assume that the higher periphyton productivity and associated higher mercury concentrations at oligotrophic U3 are solely responsible for higher mercury concentrations in the oligotrophic food chain. For clarification, the Commenter does not appear to have made the distinction between periphyton productivity and the associated concept of periphyton biomass turnover rate. While periphyton productivity is higher at eutrophic F1 than oligotrophic U3 (McCormick et al., 1998), the spatially weighted average biomass density of periphyton at U3 is such that the annual turnover of total periphyton biomass is greater at U3 than F1. Additionally, the data published by Cleckner et al. (1998b) and used in these calculations (See table 7-1 page 7-65 in ECR 2000) demonstrate that the average THg concentration in U3 periphyton was lower than at F1.

Comment: “Following the District’s reasoning, the District’s actions in removing phosphorus from the Everglades inflows will reduce macrophyte densities and concurrently increase periphyton densities in the northern parts of WCA-2A. This will spread conditions conducive to high mercury concentrations in the food webs in the ecosystem found in the south and replace the ecosystem now found in the north. One would then expect that as restoration progresses, the direct results of the District’s actions will be to increase the risk of adverse effect from Everglades wildlife from exposure to mercury. If the District wishes to stand by this position, Exponent requests that all of its ramifications, both present and future, be fully outlined in the 2001 report.”

Response: This is the basis of the reference site approach adopted by the District to predict post-ECP ecological risks to fish-eating wildlife from MeHg exposure in the restored areas of the northern Everglades. The District has explored all of the ramifications of the results of its post-ECP ecological risk assessment on pages 7-31 to 7-49 in EIR 1999 and pages 7-37 to 7-70 in ECR 2000. The District concluded that it had provided FDEP, USEPA, and the U.S. Army Corps of Engineers with the required reasonable assurance that there would not be an ecologically significant increase in post-ECP MeHg exposure to fish-eating wildlife in the restored areas of the northern Everglades. The approach, methods, data, findings, conclusion, and recommendations have been presented to the public, ECP permitting authorities, and other interested parties by inclusion in these reports. The District also addressed concerns regarding same raised in previous comments on these reports. Commenter should direct any further concerns regarding those published findings to the appropriate state and federal agencies.

- **Mercury Flux Model**

Comment: “The discussion of mercury in periphyton wrongly focuses on normalized periphyton concentrations and total mercury cycling (page 7-55). The use of mercury concentration in periphyton to discount the biodilution hypothesis is poorly grounded. The



‘coverage weighted’ values for periphyton concentrations obscure the fact that total biomass is much larger in the eutrophic zones, and thus total mercury should be more diluted.”

Response: The District’s table of data and calculation results on page 7-55 (ECR 2000) clearly demonstrates that rooted macrophyte density (g dry wt/m<sup>2</sup>) in homogeneous habitat is 1.3 times higher at eutrophic F1 than oligotrophic U3 (Miao and Sklar, 1998). Conversely, the periphyton density in homogeneous habitat at U3 is 500 times higher than F1 (McCormick et al., 1998). However, these sites are not homogeneous habitats but consist primarily of macrophyte habitat, periphyton habitat, and open water. Failure to correct for habitat coverage would result in an overestimate of both macrophyte and periphyton biomass. When habitat-normalized values are used, the calculated total biomass values for primary producers at F1 and U3 appear to be roughly equal.

Biodilution occurs when a constant source or flux of THg is diluted in an increased flux of aquatic biomass. The first manifestation of biodilution occurs in primary producers. The total plant biomass flux or turnover is calculated by multiplying the coverage, density, and productivity of each individual species and summing over all plant species. The plant-mediated flux of THg associated with total plant biomass turnover can only be calculated by multiplying the individual turnover rates for each plant species by its corresponding average THg concentration. The observed THg concentration in periphyton at U3 is lower than at F1 (Cleckner et al., 1998). In other words, the THg concentration in periphyton is more dilute at U3 than F1, converse to what would be predicted by water column TP-mediated biodilution. However, this observation is consistent with the District’s flux calculation. The District is able to explain this seemingly unexpected result based on a light limitation gradient that is maximum at eutrophic F1 and minimum at oligotrophic U3 (Grimshaw et al. (1997). Here the phenomenon of biodilution is still manifest, but it is now light-mediated rather than P-mediated.

Comment: “If we assume, as the District does, that the total mercury in periphyton mats is primarily ionic in nature, then it does in fact account for a small portion of the total spatial flux within the Everglades.”

Response: To carry out this scoping calculation, it is not necessary to assume that the THg in periphyton mats is primarily ionic (inorganic) in nature, although this is, in fact, the case (Cleckner et al., 1998b). THg is comprised of Hg(II) and MeHg.

Comment: “If the District wishes to prove this hypothesis, Exponent would like to see a demonstration of the overall methylmercury concentration in biota is lower in biomass of oligotrophic versus eutrophic sites. This is currently outside the scope of the District’s supposition because the ecological model poorly represents the productivity of the eutrophic regions of the Everglades.”

Response: Commenter has overgeneralized District’s position. To invalidate the hypothesis that TP-mediated biodilution is the predominant process regulating mercury concentrations in mosquitofish along the “F” transect, it is only necessary to demonstrate that: (1) THg turnover mediated by plant biomass turnover is higher at U3 than F1 and (2) the plant biomass weighted-average THg concentration is lower at U3 than F1. The latter is directly measurable and the former can be calculated from things that are directly measureable. The District has established (1) with the above summarized scoping calculation and (2) with empirical results. Although it is not necessary for the District to advance and defend an alternative hypothesis to invalidate the P-mediated biodilution hypothesis as applied to the “F” transect, such alternative hypotheses have been put forth (pages 7-56 to 7-62, ECR 2000).

Comment: “Aside from the error in logic and the unbalanced ecological model, there are several basic premises put forth as supportive that Exponent would like to see substantiated in the 2001 report.”

Response: The purpose of the scoping-level THg flux model was to test the hypothesis that the exponential decrease in water column TP caused an exponential increase in mosquitofish THg from eutrophic F1 to oligotrophic U3 via a decrease in plant-mediated biodilution. The scoping-level model was the least complex formulation capable of falsifying this hypothesis. The inability of this simplified model to explain the observed gradient in the magnitude of mosquitofish THg along the “F” transect does not invalidate its ability to falsify Commenter’s hypothesis. The District agrees with the Commenter that one cannot expect to predict accurately the average concentration of mosquitofish THg at F1 and U3 using an oversimplified model that omits input loading rates from EAA runoff, atmospheric deposition, and soil release, the processes of flow dilution, MeHg production and decomposition, sorption, settling, burial, and exchange, uptake and bioaccumulation, and food chain biomagnification, and the influences of water and pore water chemistries on these processes. The Everglades Mercury Cycling Model being developed for FDEP, the District, and USEPA accounts for these inputs, processes, and influences in a mechanistically realistic way. The model structure, assumptions, and calibration for application to WCA-3A-15 are summarized in Appendix 7-3 of draft ECR 2001.

Comment: “First, Grimshaw et al. (1997), the only reference provided by the District to support its arguments, only measured light availability during ‘the warmer seasons of the year.’ This would be the time of maximum Typha growth in eutrophic regions and maximum periphyton growth in the oligotrophic regions. Exponent would appreciate any evidence that this relation holds for the remaining three quarters of the year.”

Response: Grimshaw et al. (1997) measured a decrease in PAR beneath the cattail canopy in the months of March, May, and August of 1995. This spans six months of the year during which the productivity of macrophytes and periphyton is a maximum. McCormick et al. (1999) also observed this phenomenon.

Comment: “Second, overgeneralization has resulted in the District’s assumption that ionic mercury and methylmercury behave similarly throughout the environment. This is often not the case. For example, it has been shown that higher plants preferentially take up and accumulate methylmercury to a greater extent than ionic mercury (Huckabee and Blaylock 1973).”

Response: Per an earlier discussion, the District did not make the assumption that Hg(II) and MeHg behave similarly in the environment. While it is true that the District has focused on THg rather than Hg(II) and MeHg separately, it has not been the District’s experience that MeHg is a significant portion of THg in either macrophytes or periphyton. In the winter and summer of 1997, the spatially averaged arithmetic mean percent MeHg in cattail, water hyacinth, and water lettuce leaves collected from the ENR Project were 1.1% and 1.0%, 4.5% and 2.2%, and 10.1% and 2.5%, respectively (SFWMD, 1999). The percentage of the spatially averaged MeHg concentration that is THg in the ENR project interior waters averaged between 5% and 15%, so the Hg(II) BCFs for these plant tissues were generally higher than the corresponding MeHg values. This invalidates the generalization of the results of Huckabee and Blaylock (1973) to the interior of the ENR Project. Based on the unpublished results of analyses of plant tissue samples collected in July 1999 by the USGS-Madison at 3A-TH, 3A-15, 2-BS, U3, and WCA-1, the geometric mean percentages of MeHg relative to THg concentrations in sawgrass and lily pad leaves are only 2% and 6%, respectively. This invalidates the generalization of the results of Huckabee and Blaylock (1973) to the interior Everglades.

Comment: “If we assume, as the District does, that the ‘total mercury’ in the macrophytes is ionic in nature, then it does in fact account for a small portion of the total spatial flux within the Everglades.”

Response: See previous response.

Comment: “However, if the measured total mercury concentration in the macrophytes represents disproportionately higher concentrations of methylmercury, it may well represent a highly significant pathway for the introduction of methylmercury into the food chains.”

Response: See previous responses on this topic and the related topic of the inverse relationship with TP.

Comment: “Therefore, Exponent requests that in the 2001 report the District provide the following:

Consideration of the relationship between productivity and nutrient balance for the entire year, not just a small part of the year.

The data on which the analysis in Table 7-1 was performed.

The distributions from both the eutrophic and oligotrophic regions (rather than generalizing thousands of acres with a single number).

Analysis of ionic mercury and methylmercury separately in their models, to test this flux hypothesis.”

Response: The periphyton production data from McCormick et al (1998, 1999, 2000) were collected year around, rather than just in the spring and summer, as was the case with the light-limitation data collected by Grimshaw et al. (1997).

The majority of the data are available from the open literature. Unpublished data used in the calculations are presented in Table 7-1 on page 7-55 in ECR (2000).

The spreadsheet contains all of the relevant data on coverages, densities, and production rates measured at F1 and U3. Generalization from representative quadrats to the scale of thousands of acres is necessitated by the District’s inability to measure these values for 4,047,0000 1 m<sup>2</sup> quadrats in 1,000 acres.

This has been carried out and provides further support for the District’s conclusion that water column P-limited biodilution is not the cause of the increase in mosquitofish THg with downstream distance along the “F” transect in WCA-2A. See comments above.

- **Relationship of Mercury to Sulfur Cycling**

Comment: “Heavy reliance on personal communications appears again in the District’s discussion of the role of sulfate in mercury cycling. For example, the observation that the absence of sulfate ‘is not always associated with inactivity of SRB’ is key to the discussion of methylation, yet is supported only by informal communication from Cindy Gilmour.”

Response: The District does not understand why this observation, which was included for the sake of completeness, is “key to the discussion of methylation.” To the contrary, the District believes that this is of no particular significance to understanding the methylation of Hg(II) in the northern and central Everglades, where sulfate is always in sufficient supply to prevent SRB switchover to fermentation.

Comment: “More importantly, the reference to demethylation by aerobic bacteria, SRB, and methanogens is probably the most important component in understanding net methylation rates; yet the only citation is a personal communication from R. Orm (sic), USGS. Exponent again requests that if peer-reviews evidence is not available to substantiate the District’s arguments, then the District present data upon which these premises are based in the 2001 report.”

Response: The paper by Marvin-DiPasquale and Oremland (1998) cited on page 7-57 of ECR 2000 reports the results of measurement of the rates of demethylation by methanogens and SRB using radiolabelled MeHg in sediment cores obtained from a number of Everglades sites.

Comment: “Overall, Exponent feels that the validity of the sulfur hypothesis is tenuous at best.”

Response: Consistent with the protocols of scientific discourse, the District urges Exponent to bring its objections to the attention of the editors of Biogeochemistry regarding Gilmour et al. (1998a) and Cleckner et al. (1998a,b), Environmental Science and Technology regarding Marvin-DiPasquale and Oremland (1998) and Jay et al. (2000), the Journal of Environmental Chemistry and Toxicology regarding Benoit et al. (1998a,b, 1999), and Limnology and Oceanography regarding Cleckner et al. (1999) to address the tenuousness of the sulfur hypothesis. This is not the proper forum in which to challenge such published results.

Comment: “The discussions regarding mercury methylation and demethylation are not connected, as they must be because net methylation is the principal parameter that correlates with food web concentrations.”

Response: When read in context, the required connections are made. However, while soil sampling for these measurements occurs at the same sites and times, it is the measurements of methylation and demethylation routes and rates themselves that are disconnected. The District has rectified this situation by contracting with Mark Marvin-DiPasquale, Ph.D., at USGS-Menlo Park to carry-out one-flask studies of the effects of various factors on gross methylation and gross demethylation simultaneously. The District does agree with Commenter that net methylation, not P-mediated biodilution, is the most important determinant of MeHg concentrations in the food web. As a consequence, the District has determined to cease its biodilution studies along the WCA-2A “F” transect in August 2000. The work of Krabbenhoft and others, Gilmour and others, and Marvin-DiPasquale and others will continue to ensure that our mechanistic understanding and predictive modeling capability will continue to support well-informed resource management decision-making.

Comment: “For example, the discussion on the bottom of page 7-57 takes great care to describe the parabolic relationship between sulfate concentration and net methylmercury production. However, this is based on what is described as a ‘moderate inverse relationship’ between sulfate concentration and methylmercury in soil.”

Response: Commenter has not quoted the sentence correctly and has taken this statement out of context. As written, the sentence reads “As depicted in Figures 7-13 and 7-14, there appears to be a moderate inverse correlation of methylmercury in Everglades soils with peat pore water

sulfate, but a much stronger inverse correlation with peat pore water sulfide (C. Gilmour, ANS, pers. comm., 1999).” The observation that MeHg production increases as pore water sulfate increases up to a point and is then suppressed in increasingly sulfidic environments is not new, but the earlier hypothesis that this is caused by the formation of HgS (cinnabar) precipitate has been discounted as an oversimplification. In fact, Ravichadran et al. (1998) demonstrated that Everglades DOC could prevent the precipitation of and dissolve precipitated HgS under conditions of temperature, pH, and ionicity in which it was predicted to be present as a stable precipitate based on thermochemical first principles. The new hypothesis proposes a mechanism involving the formation of charged and uncharged sulfide complexes of Hg(II), with the uncharged complex readily passing through SRB membranes via passive diffusion, while the charged species cannot (Benoit et al., 1999a,b; Jay et al., 2000).

The sulfur hypothesis is now undergoing further testing in the laboratory and in the field using a suite of stable isotopes of mercury and a radioisotope of sulfur. The sulfur hypothesis and the proposed studies to test and refine it are summarized by Gilmour and Krabbenhoft in some detail in Appendix 7-4 of draft ECR 2001.

Comment: “The District expresses confidence that the peak net methylation rate, as well as mercury concentrations in sediment, water, and biota, can be predicted from the optimal ratio of sulfate to sulfide in pore water. However, while the high methylation rates at WCA-2B-S might be explained by sulfate/sulfide ratios, this does not explain why other regions of the Everglades with similar sulfate/sulfide ratios do not show the same levels of methylmercury production and bioaccumulation (see page 7-59). The relationship at WCA-2B-S appears to be correlation, not causation. Furthermore, the District’s supposition that groundwater flow, or wetting and drying cycles, or mercury deposition rates may override the importance of the sulfate/sulfide ratios only emphasizes the weakness in the model (i.e., it has failed to capture the key environmental factors involved in the regulation of net mercury methylation.)”

Response: It is not the District’s position that the pore water sulfide/sulfate ratio controls MeHg production and bioaccumulation under all circumstances. However, all other factors being equal, the pore water sulfide/sulfate ratio should be a strong predictor of MeHg production, and, taking into account hydrological and seasonal influences on trophic dynamics, of MeHg bioaccumulation, as well. The discussion of examples of areas where the magnitude of MeHg bioaccumulation does not appear to be controlled by the pore water sulfide/sulfate ratio was included to underscore the importance of not relying on correlation to infer causation and the concomitant importance of carrying out mechanistic studies under controlled conditions to test hypotheses generated by observational studies. The mechanistic model being developed by Tetra Tech for the District and FDEP does capture all of the key transport, biogeochemical, and bioaccumulation processes in a mechanistically realistic, self-consistent way to guide well-informed restoration management decision-making. In particular, the model will be used to predict the change in methylmercury bioaccumulation over time in response to various combinations of load reduction, water quantity changes, and water quality changes, including the benefits of the anticipated post-ECP reduction in the mercury load in Lake Okeechobee releases and EAA runoff.

### **Probabilistic Risk Assessment (Appendix 7-3b)**

Comment: “because the District did not follow through on the appropriate analyses, there is no way to validate their accuracy, and therefore, their estimates cannot be accepted.”

Response: After careful consideration of each issue the Commenter raised, the District finds no significant error in the application or in the reporting of the results of its probabilistic ecological risk assessment (PERA). The District followed USEPA guidance and standard methods in performing and reporting its PERA. Moreover, the District went beyond these minimum requirements to validate its exposure assumptions and model. When this was done, the District's exposure model was shown to accurately predict tissue concentrations in egret eggs and feathers. Furthermore, the District calls to the Commenter's attention the fact that it did not rely solely on a mathematical exposure model in its assessment of risk, but instead made its determination based on multiple lines of evidence. These other lines of evidence included the results of epidemiological and controlled dosing studies of great egrets and other wading birds. Finally, with the recent revision to Exponent's risk assessment (MacKay et al., 2000), risk estimates have now converged, and the remaining differences between Exponent's assessment and the District's assessment appear to center more on interpretation and communication of risk.

Comment: The Commenter challenges the District's documentation of its methods and states there are references cited in the text that are not provided in the reference section. The Commenter also recommends that the District consult EPA's draft probabilistic guidance document (USEPA, 1999).

Response: The author of the District's PERA is familiar with the guidance document (USEPA, 1999), and recently participated in a workshop, sponsored by USEPA and the National Institute for Environmental Health Sciences, where the recommendations were presented by Dr. S. Chang (a principal USEPA contributor to the document). The District's PERA was presented as a poster at this same workshop (Rumbold, 2000b).

The documentation of methods used in the District's assessment was deemed appropriate for a 1-dimensional Monte Carlo Analysis (1-D MCA) when evaluated using a checklist produced for reviewers of USEPA assessments (Table 6-1; USEPA, 1999) and when compared to recently published and unpublished risk assessments (Hope, 1999; Sample and Suter, 1999; Mackay et al., 2000). Specifically, the District's risk assessment contained a conceptual model, explicit descriptions of assessment endpoints, data sources, parameter values, distribution forms, information on truncations, concentration terms, algorithms, and methodology for estimation of tail stability (i.e., iterations were performed until the between-iteration change in percentile was below 1.5 percent). Moreover, in accordance with the guidance document (section 5.3.1, USEPA, 1999), graphical presentation of the results showed both the estimated NOAEL-based risk and the LOAEL-based risk (cf. Mackay et al., 2000).

Nevertheless, the Commenter's specific concern about insufficient documentation of the normalization procedure was valid. As outlined in the District's response to comments submitted by the Commenter on 11/05/99 and 11/07/99 (pages A1-3-61 to 96; SFWMD, 2000), the District relied on a simple, commonly used, two-step procedure built into the model that automatically normalized the percent diet fraction so that the sum equaled 100%. The Appendix should have documented that procedure (this issue is discussed further below in another response).

With regard to references, after cross checking the 53 references in the literature cited section with references cited in the document, the District found three errors: one missing reference and two errors in date of publication – see below. The missing reference is as follows: Kahl, M.P. 1964. Food ecology of the wood stork (*Mycteria americana*) in Florida. Ecol. Monogr. 34: 97-117.

Date errors were: "Kahl, 1963" cited in footnote to Table 1 should read Kahl, 1964; "Gawlik, D.E. (ed.) 1990", should read Gawlik, D.E. (ed.) 1999.

- **Lack of Distribution Testing**

Comment: The Commenter challenges the District's use of parametric distribution functions (PDFs) and wishes to see results of goodness-of-fit analyses.

Response: The District has previously responded to similar comments submitted by the Commenter on 11/05/99 and 11/07/99 (pages A1-3-61 to 96; SFWMD, 2000). The District would like to expand on its response by referring the Commenter back to the USEPA guidance document (1999); specifically the Commenter is referred to section 3.4 Fitting Distribution to data, section 3.4.2 Empirical Distribution Functions (EDFs) and section 3.5.3 Cautions Regarding Goodness-of-fit tests.

Use of both PDFs and EDFs are based on the assumption of random, representative data sets. A strictly empirical approach does not allow extrapolation beyond the range of observed data. Consequently, EDFs may not adequately represent the tails of the distribution due to limitations in the number of samples collected or in sampling methods. Moreover, the shape of the EDF may be attributable to random fluctuations or sampling bias. Accordingly, for its risk assessment, the District developed and evaluated PDFs using three different goodness-of-fit tests: the chi-square test, Kolmogorov-Smirnov and, Anderson-Darling goodness-of-fit tests (see documentation for Crystal Ball® software). However, due to the limitations of these tests (see section 3.5.3 – USEPA, 1999), the District also relied heavily on graphical analysis (USEPA, 1999: “In addition, the value of graphical evaluations of the fit cannot be overstated”), where appropriate, defaulted to common parametric distributions (e.g., lognormal for tissue concentrations), and relied on the published literature (e.g., Henning et al., 1999, etc.).

However, the District was concerned that the sampled fish population collected via electroshocking may not have been representative of a wading bird's prey population. This concern was indirectly raised also by the Commenter (comments 11/05/99 and 11/07/99), when he pointed out that fish collected as regurgitant of the great egret must be considered the most direct measure of the receptor's actual exposure. Electroshocking and netting tends to result in samples biased toward larger fish. Accordingly, the District devoted its resources to testing the validity of the exposure model by predicting the resulting residue concentrations in eggs and feathers and comparing these predictions with observed concentrations (see Figure 7-3b-6 and Table 2; Rumbold, 2000a). As previously mentioned, predicted residue levels were higher than observed concentrations in eggs and feathers, suggesting that the model overestimated exposure, at least for the great egret.

- **Improper Model Execution**

Comment: The Commenter challenges the District's model, and in particular the normalization of prey fractions (also see comment on summations of probability made in Mackay et al., 2000).

Response: The District has already responded to similar comments that the Commenter submitted on 11/05/99 and 11/07/99 (for response see pages A1-3-61 to 96; SFWMD, 2000). The District reiterates that it used a commonly accepted modification of the generalized exposure model and refers the Commenter to refereed papers by Sample and Suter (1999) and Hope (1999)(also see earlier response regarding documentation of the model). In their assessment of methylmercury and PCB risks at the Oak Ridge Reservation, Sample and Suter (1999) used the following exposure model:

$$E_j = \left( \frac{IR_w \times CW_j}{BW} \right) + \left( P_a \times \sum_{i=1}^n \frac{(P_i \times IR_f) \times CF_{ij}}{BW} \right)$$

Where  $E_j$  = exposure to contaminant (j; mg kg<sup>-1</sup> day<sup>-1</sup>),  $IR_w$  = ingestion rate of water (liters day<sup>-1</sup>),  $CW_j$  = concentration of contaminant (j) in water (mg l<sup>-1</sup>),  $BW$  = body weight (kg),  $P_a$  = proportion of fish or other aquatic prey in diet,  $n$  = number of size categories of fish consumed,  $P_i$  = proportion of fish in diet in size class  $i$  (mg kg<sup>-1</sup>),  $IR_f$  = ingestion rate of food (kg day<sup>-1</sup>),  $CF_{ij}$  = concentration of contaminant (j) in fish in size class  $i$  (mg kg<sup>-1</sup>).

Except for the absence of an exposure term from water ingestion, which is not a risk driver in the Everglades, and the focus on prey species rather than prey size, the District's model (Rumbold, 2000a; pg. A7-3b-7) is nearly equivalent to Sample and Suter's model. The other difference is that Sample and Suter (1999) fixed the proportion of fish in each of the size classes (i.e., the  $P_i$ s were point estimates) and, thus, this term did not require normalization. However, other assessments that do allow prey fractions to vary often include a normalization step identical to that used in the District's model. For example, Hope (1999, pg. 155) states:

"To prevent the sum of  $F_{ij}$  for all dietary items for a given consumer from exceeding 1 during any iteration of the model, the individual  $F_{ij}$  values were normalized relative to their sum, as follows:

$$Fn_{ij} = \frac{F_{ij}}{\sum_{i=1} F_{ij}}$$

Where:  $F_{ij}$  = Dietary fraction of the  $j$ th food item in the diet of the  $i$ th receptor, unitless

$Fn_{ij}$  = Normalized dietary fraction of the  $j$ th food item in diet of the  $i$ th receptor, unitless.

(Note: Bruce Hope is the current Chair of the Ecological Risk Assessment Specialty Group of the Society for Risk Analysis)

While exposures models can be useful, some degree of model verification is important to reduce uncertainty (cf. MacKay et al., 2000). The Commenter is referred back to Figure 6 and Table 2 of Appendix 7-3b (Rumbold, 2000a) that compared model-predicted concentrations with concentrations observed in collected eggs and feathers. Based on these comparisons, the District's model was found to be conservative, i.e., in that it overestimated mercury exposure to the receptor population.

- **Lack of consideration for correlation**

Comment: The Commenter raises three different issues under this subheading regarding correlation:



“... the District’s model explicitly requires that fish size be independent of fish type”;

“... the District neglected the correlation, noted by Krebs (1974), that larger herons tend to eat larger fish”; and

“... the District also overlooked the correlation between heron size and species of fish eaten”.

Response: While it would be preferable to parameterize preferred prey size within each prey species and to model the correlation between tissue mercury concentration and prey size (i.e., analogous to a nested effect), this would require huge data sets, which are most often lacking. Instead, diet is typically modeled in one of two ways. One approach is to ignore prey species and construct a model based solely on preferred prey size. The second approach is to model diet based on preferred prey species and ignore prey size. The previously cited risk assessment by Sample and Suter (1999) is an example of the former approach. In performing its assessment, the District compared the two different approaches and found the model based on prey species was more conservative, at least for great blue heron and great egrets. In contrast, wood storks had slightly higher exposure if they took fish solely based on preferred size. The District chose to model diet based on preferred prey species (only setting an upper bound prey size) because: (1) of the availability of data from the published literature to model prey selection based on species, (2) the prey species approach was more conservative for the great blue heron and great egret, and (3) work by Ogden et al. (1976) suggests the wood stork selects prey based on species (i.e., due to prey’s behavior, density or vertical distribution).

The Commenter's second and third comments will be addressed together. Clearly, different species of herons that are of different size vary in their preferred prey species and prey size. However, after careful review of the reference cited by the Commenter (i.e., Krebs, 1974), the District found no data or observation that herons changed prey species or size preference as a function of bird size. Furthermore, in his review of wading bird feeding ecology, Kushlan (1978) presented no evidence that supports either a change in prey size or prey composition in different sized herons of a given species. To the contrary, both Kushlan (1978) and Kahl (1964) report that even nestlings are offered the same prey as taken by adults.

- **Lack of Confidence Limit Reporting**

Comment: The Commenter challenges the District’s use of a “first-order” (i.e., 1-dimensional) Monte Carlo analysis rather than a “second-order model” (i.e., 2 Dimensional).

Response: The Commenter is referred to Figure 1.4 in the USEPA guidance document (1999) that shows the tiered approach advocated by the agency for conducting risk assessments. From Figure 1.4, it should be clear that the District has responded appropriately at each “scientific/management decision point” in conducting its risk assessment, beginning first with a deterministic risk assessment in 1998 (Rumbold et al., 1999) and then progressing to a 1-dimensional Monte Carlo Analysis (Rumbold, 2000a). According to the guidance document, the results of this initial PRA are to be evaluated to determine if the analysis should progress to tier 3. The first question that should be addressed is whether or not the risk estimates from both the deterministic risk assessment (i.e., based on point estimates) and the 1-D MCA are of concern for the potentially exposed population (for a review of the District’s conclusions, see Rumbold, 2000a).

Equally important, USEPA (1999) recommends that the results of the initial probabilistic simulation be the centerpiece of the workplan for the next tier of the PRA. For example,

sensitivity analyses carried out during the District's assessment identified specific data gaps (e.g., tissue concentrations in: small sized sunfish and bass, warmouth at certain sites, trophic level 1 and 2 fish) that should be filled before proceeding to the next tier. Nevertheless, it is unlikely that the basic conclusions drawn here will change when these data gaps are filled and another iteration is performed.

- **Inappropriate Regional Comparisons**

Comment: The Commenter requests that the District assess the risks at WCA-2A F1.

Response: The criteria for selecting WCA-2 U3 and WCA-3A 15 as the Post-ECP reference site and positive control site, respectively, have been reviewed previously (Rumbold et al., 1999; Rumbold, 2000a). For its 1998 deterministic risk assessment, the District relied on the Florida Game and Freshwater Fish Commission data set. At that time, large-bodied fish were not routinely sampled at WCA-2A F1. Subsequently, the Game Commission (now the Florida Fish and Wildlife Conservation Commission), under contract to the District, has conducted two annual collections at WCA-2A F1 (using methods identical to those used to sample other sites). However, no large-bodied fish (e.g., sunfish or bass) were collected during either sampling event. According to the FFWCC contractor, this was "due to a lack of habitat at F1" (Lange et al., 1999).

Clearly, it would be informative to estimate the current risk at F1. However, this is not as critical as estimating the future risk at F1 following water quality improvements, which should improve the fish habitat. It is critical to estimate future risks at F1 because, contrary to the Commenter's next comment, the District fully appreciates that conditions at F1 will change when phosphorus is reduced to U3-like levels. Fortunately, future risk at F1 can be estimated using the extensive data set from the U3 site.

- **Improper Result Interpretations**

Comment: "Exponent is concerned that a population of wildlife receptor, such as the great blue heron, that possesses a 3-year maturation cycle cannot endure this level of reproductive failure without requiring recruitment from the outside population."

Response: (From the preceding statement, the District assumes the Commenter is suggesting that one-fifth of the great blue heron population will suffer reproductive failure).

With the recent revision to Exponent's risk assessment and the reduction in their hazard quotient, which has decreased from about 10 to about 1.5 for the median exposed great blue heron (i.e., 50<sup>th</sup> percentile; Exponent, 1998; MacKay et al., 2000), the principle differences remaining between Exponent's assessment and the District's assessment are interpretation and communication of risk. To some degree this is understandable, because there are no ecological risk levels that are recognized as being either clearly acceptable (*de minimis*) or clearly unacceptable (*de manifestis*) (Suter, 1993). The USEPA guidance document (1999, pg. 5-35) states:

"In contrast to the case for human health risk assessments (where default risk-based decision guidelines are well established), there are no established default decision guidelines for identifying when risks to ecological receptors are of concern. However, if risks to nearly all members of the population (including the RME individual) are below the appropriate NOAEL-based toxicity reference value, then it is likely that risks are within an acceptable range both for

individuals and the population. Conversely, if risks to the average members of a population exceed an appropriate LOAEL-based TRV, then it is likely that risks are not acceptable, either to individuals or the population.”

The District’s probabilistic risk assessment predicted 78.2% of the modeled great blue heron population at the Post-ECP reference site would receive a daily dose rate below the estimated no-observed-adverse-effect level (NOAEL, Fig. 1). Hence, this segment of the population is believed to have no appreciable risk of deleterious effects from mercury exposures.

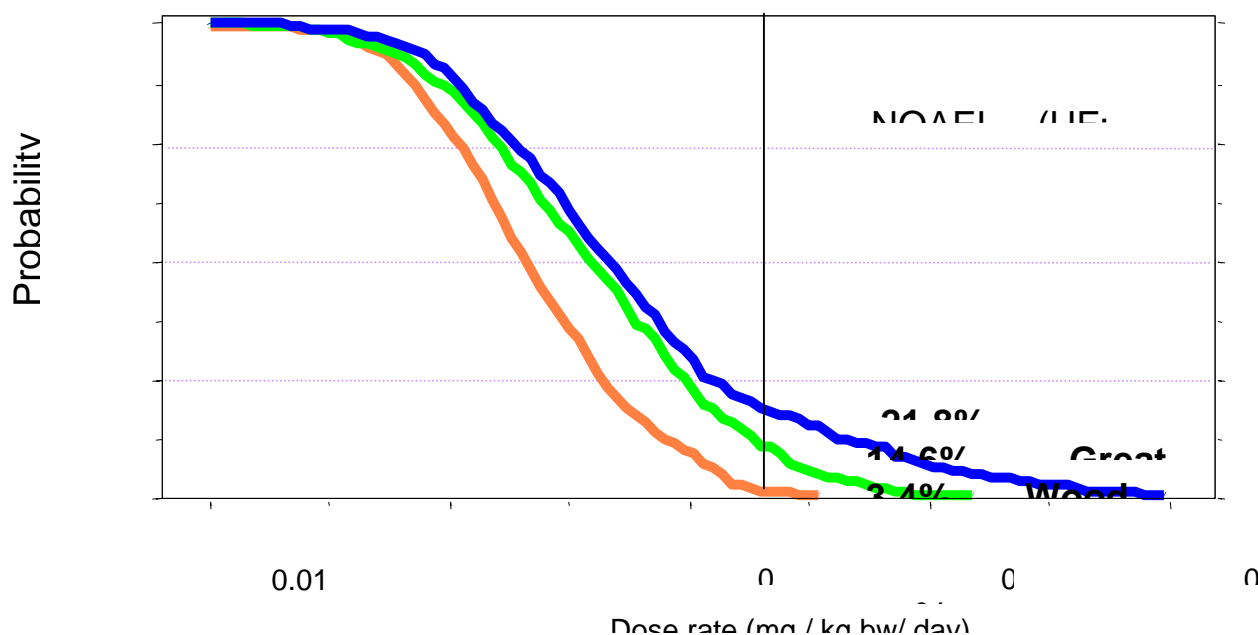


Figure 1. Reverse cumulative density function of dose rate for mercury exposure to wading birds feeding exclusively at the Post-ECP reference site (Fig. 7; Rumbold, 2000). Note, the lowest-observed-adverse-effect level (LOAEL), 0.064 mg/kg-bw/day, not shown on x-axis.

However, Figure 1 indicates also that 21.8% of the population is expected to receive a daily dose rate exceeding the NOAEL and, thus, are at some elevated risk. That is not to say that 21.8% of the population will suffer reproductive failure.

To assess the biological significance of the risk to this great blue heron population, one must first understand the basis of the LOAEL and the derived NOAEL-based toxicity reference value (TRV) that was used to evaluate the exposure distribution. This includes the nature of the LOAEL endpoint, its relevance to the assessment endpoint, and the shape (steepness) of the dose-response curve (USEPA, 1999).

As discussed in Appendix 7.3b (Rumbold, 2000a), the USEPA derived the LOAEL from a series of studies carried out by Gary Heinz (USEPA, 1993; 1995). Heinz dosed three generations of mallard duck hens ( $n = 74$ ) at 0.5 mg MeHg/kg food eaten. During the first generation study, Heinz (1979) determined that 0.5 mg MeHg/kg food had no significant effect on reproductive effects such as hatching success or duckling survival (i.e., no-observed-adverse-effect-level or

NOAEL; dose rate of @ 0.064 mg/kg-bw/day; see Rumbold, 2000a regarding calculation). However, second generation dosed birds (and when data were pooled from all three generations) showed significant differences from controls in terms of:

- Percent of eggs laid outside nestbox
- Percent of sound eggs
- Number of 1-week-old nestlings produced
- Percent of ducklings approaching calls
- Distance traveled in avoidance

Because the study included only a single dosing level, Heinz (1979) did not develop a dose-response curve for any of the measured effects. While the differences between the dosed and control birds were statistically significant, the size of the average effect was often small. For example, in terms of mean measurements from the three generations combined:

- Percent of eggs laid outside the nestbox increased from 4.3% in the controls to 9.7% in the dosed group.
- Percent of sound eggs decreased from 0.69 to 0.59 eggs laid per hen per day in the dosed group.
- Number of 1-week-old nestlings produced decreased from 46 in the controls to 37.5 in the dosed group.
- Percent of ducklings approaching calls decreased from 97% in the controls to 94% in the dosed group.

Distance traveled in avoidance increased from 32 cm in the controls to 38 cm in the dosed group.

While it is clear that some of the mallards suffered decreased reproductive success, the mallards did not suffer complete reproductive failure when dosed at the LOAEL. Accordingly, it would be incorrect to infer that 21.8% of the great blue heron population at the Post-ECP reference site will suffer reproductive failure because their exposure exceeded the estimated NOAEL, which is ½ the dose rate of the mallard LOAEL (cf. MacKay et al., 2000). Instead, the only conclusion that can be drawn from the available data is that these birds are at elevated risk of some adverse effect.

The District's assessment attempted to provide perspective for assessing this risk using a positive control site, i.e., the mercury "Hot Spot" at WCA-3A 15. Despite a much higher risk level (i.e., > 99% exceeded NOAEL and 83.8% exceeded LOAEL; see Fig. 8 – Rumbold, 2000a), no evidence of impaired reproduction (i.e., fledging success) has been reported for wading birds nesting near the positive control site (for review see Rumbold, 2000a). In fact, quite the opposite has been reported. The 3-year running average for great egret nests in the Everglades has increased since the mid-1980s and peaked in 1999 (1999 nesting approached record nesting observed in 1940s; Ogden, 1999). Further, clutch size and more importantly nest success for great egret colonies in WCA-3A have recently been the highest recorded for the Everglades and in the upper range for the species, overall (Frederick et al., 1997). As previously mentioned, the

District also validated its exposure model by predicting concentrations of mercury in great egret eggs and feathers collected from colonies near the positive control site. The results of the validation exercise showed that levels observed in eggs and feathers were less than model-predicted concentrations and suggest that the 1-D MCA overestimated mercury exposure to the birds. More importantly, observed tissue concentrations near the “Hot Spot” were also below levels thought to be of toxicological concern (Rumbold, 2000c).

As evident from this discussion, in accordance with USEPA guidelines for ecological risk assessment (1998), the District did not rely solely on a mathematical exposure model in its assessment of risk at WCA-2A U3, but instead made its determination based on multiple lines of evidence (cf. MacKay et al., 2000).

### **Historical Trends of Mercury Concentrations in Everglades Fish and the Feathers and Eggs of Wading Birds**

Comment: 1. “Exponent remains unconvinced that the reported observations are real because the District has not performed the appropriate statistical analysis on the data sets provided by Frederick et al. (1997) and Lange et al. (1993).”

Comment: 2. “Exponent analyzed the same data sets used in the 2000 report and demonstrated, based on multiple Studentized t-tests, that there was no significant difference with time in the concentrations of mercury in the prey fish for the wading bird receptors.”

Comment: 3. “we [Exponent] could not perform the same analysis on the egg and feather data because the District failed to provide the data in a timely manner.”

He later requests:

Comment: 4. “the District to include in the 2001 report a proper hypothesis testing with rigorous statistical analysis of these data sets. These should include factorial analysis of variance to ensure the differences in the mean represent a true trend, as well as covariance analysis to ensure that circumstances such as time of year of collection, size of fish, or location of nesting sites have not biased the results.”

Response: Before addressing the specific comments, it is important to note that the District did not rely on a projected decreasing concentration trend in fish as mitigative of long-term exposure to estimate risk in either its deterministic or probabilistic models. The District modeled exposure based on mercury concentrations observed in fish collected at the Post-ECP reference site and positive control site from 1997 through early 1999.

With regard to issues 1 and 4, the District refers the Commenter to Chapter 7 and to appendix A7-9 of the draft ECR 2001. The latter has a thorough discussion of the statistical methods, which included ANOVA and ANCOVA, that were used in the analyses of data sets collected by the District. The period of record of the District’s data sets continue to expand and has already proven invaluable in assessing temporal and spatial trends in mercury bioaccumulation. However, as pointed out by the Commenter, two of the most extensive data sets were, and continue to be, collected by the Florida Fish and Wildlife Conservation Commission (FFWCC - Lange et al.) and by researchers from University of Florida (Frederick et al.). Both the ECR 2000 and the draft ECR 2001 cite work done by these two entities that suggest mercury concentrations have declined in fish and birds over the last 3-5 years.

Because these programs are on-going, the District does not feel it necessary to carry out independent analyses of these data sets. Moreover, as illustrated by the Commenter's comment regarding the public records request (and a previous request for data sets dealing with the inverse relationship between phosphorus and mercury), if the District were to carry out such analyses, the District would have to be free to provide the raw data upon request. The District feels it more appropriate that the two entities responsible for study design and implementation continue to independently analyze (and disseminate) their own data as appropriate. As cited in Appendix A7-9 (Rumbold, in review), results from both programs were recently presented at the South Florida Mercury Science Program's All Investigators Meeting (Lange et al. 2000; Frederick and Spalding, 2000; Palm Harbor, Florida. May 9-11, 2000). The Commenter was in attendance at this meeting.

The second issue the Commenter raises is subsequently addressed in his 4<sup>th</sup> comment. Fish size is typically a significant covariate to tissue mercury concentration. Consequently, if sampled populations differed in size, a simple t-test would be an inappropriate statistical method to assess differences in tissue concentrations. Analysis of covariance (ANCOVA) would be the more appropriate statistical test (for details see Appendix A7-9). The FFWCC has performed ANCOVA on the mercury concentrations in bass fillets and reported a significant downward trend over time at certain locations (for details see Lange et al., 1999; 2000). Regarding the other covariates identified by the Commenter, both time of year and collection sites have been standardized to remove these sources of variability.

With regard to item 3, while the District did receive and fulfilled public record requests from Exponent for certain internal documents and raw data on mercury concentrations in fish tissues, it never received a request, either formal or informal, for data on mercury concentrations in eggs or feathers. Further, results of the District's analyses of these data sets, which included arithmetic means, standardized least square means, and variances, were reported in Appendix 7-4 in the draft ECR 2000 that was available to the public on September 7, 1999 (published in the final document as appendix 7-3a on January 1, 2000). Pursuant to Florida's public records law, it is the policy of the District to fulfill all public record requests.

To facilitate any such future requests, the Commenter is referred to Donna Kramer in Government Affairs and Communications. She can be reached at (561) 682-6215.

Comment: Specific requests for inclusion in the 2001 report include:

"Details on the design of pharmacokinetics models that the District will use to determine the impact of its activities on the Florida panther, Everglades mink, and river otter."

Response: For purposes of developing a revised water quality criterion for total mercury (THg) as methylmercury (MeHg) to protect species indigenous to South Florida, it is appropriate to evaluate the dietary exposure rates and toxicological sensitivities of the Everglades mink (*Mustela vison Evergladensis* Hamilton), the Florida panther (*Felis concolor coryii*), and the river otter (*Lutra canadensis*). However, for purposes of evaluating post-ECP mercury impacts in the areas of the northern and central Everglades already impacted by excess nutrients, it is only appropriate to protect the most exposed, most sensitive fish-eating species and their predators indigenous to those areas. The endangered Everglades mink was originally indigenous to a highly restricted range to the coastal area near Everglades in Collier County and exhibited a preference for mangrove and cypress marsh habitats (Burt, 1949). Although development (e.g., dirt roads and levees) in the area of its original range may have allowed some expansion (Humphrey, 1992), the Everglades mink is not now indigenous to the areas of the northern and central Everglades that will be affected by post-ECP changes in water quality or quantity. This was noted on page

7-26 of ECR 2000. This is also the case for the Florida panther (*Felis concolor coryii*), whose preferred range excludes the impoundments of the northern and central Everglades (FPIC, 1989). Therefore, the Everglades mink and the Florida panther should be given no further consideration as potentially highly exposed receptor species of concern for purposes of post-ECP mercury impact assessment. This should not be the case, however, when revising the numerical Class III water quality criterion for THg as MeHg to protect highly exposed fish-eating animals and their predators, including terrestrial mammals.

Rather than awaiting the toxicological results of long-term feeding studies of the otter and the Florida panther (or the Texas cougar as its non-endangered surrogate), the revised water quality criterion for THg is likely to be derived via interspecies extrapolation of exposure and effects data from the MeHg toxicology literature for the domestic cat (*Felis domesticus*) to the Florida panther and for farm-raised mink (*Mustela vison* mink) to the river otter (*Lutra canadensis*). The purpose of the proposed pharmacokinetics studies is to reduce the greatest sources of uncertainty in these inter-species extrapolations in the shortest timeframe in the most cost-effective manner. The rationale, objectives, and general approach of the pharmacokinetics studies for the panther, otter, and mink are described on page 7-74 of ECR 2000. However, while the District and FDEP are required to evaluate the protectiveness of the existing THg WQS, FDEP has the lead in designing, implementing, and interpreting and applying the results of the studies required to derive a revised numerical WQS for THg more protective of humans and wildlife than the existing numerical Class III WQS.

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[To Be Supplied]

## Chapter 7: The Everglades Mercury Problem

### Response to Comments from Gary Bigham of Exponent on behalf of the Sugar Cane Growers Cooperative submitted August, 18 2000

Prepared By: Darren Rumbold

During the preparation of the Everglades Consolidated Report (ECR) 2001, the District received a technical memorandum from Gary Bigham of Exponent on behalf of the Sugar Cane Growers Cooperative (dated August 18, 2000) entitled “*Evaluation of Population Risks to Avian and Mammalian Wildlife in the Northern Everglades.*” After careful review of the information contained within this document, the District concluded that risk estimates have converged, and the remaining differences between Exponent’s assessment and the District’s assessment (Appendix 7-3b, ECR 2000) appear to center primarily on interpretation and communication of risk (for additional discussion, see District’s responses to Mr. Bigham’s March 31, 2000 comments; also published as part of this appendix). This convergence of risk estimates is not surprising because the period-of-record of fish contamination data sets (Lange et al. 1998, 1999) that the two assessments used to quantify methylmercury exposures overlapped. Nevertheless, to ensure that Exponent’s assessment was given proper consideration, it was forwarded, along with a copy of Appendix 7-3b of the ECR 2000 report, to the Peer Review Panel on September 12, 2000. While the Panel did not comment directly on Exponent’s assessment, except for a preliminary comment by Dr. Kent (posted to the webboard on September 23<sup>rd</sup>, 2000), in their final report they did conclude that:

“The SFWMD is to be commended for their use of probabilistic risk assessment for the wading birds. This is the current cutting edge risk methodology, and the assumptions and parameters used in these assessments are sound and reasonable. While the NOEL may be high because of its development for Mallards, it is the conservative approach until another NOEL can be developed that is more appropriate. A sensitivity analysis may indicate what factors are most subject to having an effect on the affects endpoints.”

## CHAPTER 7: THE EVERGLADES MERCURY PROBLEM

### Responses to Questions Asked Prior to the October 6, Peer Review Panel Workshop by Dr. Joanna Burger.

By Thomas Atkeson & Paul Parks (DEP) and Larry Fink  
and Darren Rumbold (SFWMD)

#### OVERALL

It was an excellent idea to organize the chapter with introductory material followed by summaries of all the major points concerning the mercury problem, with appropriate research. The in-depth appendices that followed were then much easier to follow and provided the details that were essential.

I have a number of overall questions, as well as questions from individual sections. General questions follow:

Comment: Both the bioavailability and assimilation of mercury are dependent upon the presence of a number of other elements, such as selenium, which decreases uptake and assimilation in vertebrates. It would be useful to know the degree to which this is taken into account in various calculations, and to know the levels of selenium in the different STAs.

Response: Selenium may mediate methylmercury (MeHg) bioaccumulation and toxicity, but monitoring routinely for it in water, fish, vegetation, and sediment would have more rapidly depleted already limited resources than could be justified based on the likely magnitude of its influences and the likely differences in its concentrations across the Everglades. Thus, selenium and other trace elements have not yet been taken into account in modeling methylmercury production and bioaccumulation. Our strategy is to discover those processes governing the net rates of mercury methylation and bioaccumulation and the physical, chemical, and biological factors that influence those processes. This will be followed by an attempt to determine if any potentially controllable factors affect the net rate of methylation. It might be helpful to know how other factors affect bioavailability and assimilation at any given level of available methylmercury, but the complex relation between food web dynamics and factors governing the net rate of methylmercury production and bioaccumulation make the interpretation of the results of such correlative studies problematic. Also, factors affecting bioavailability and assimilation would be of importance in alleviating mercury bioaccumulation only if there were some potential for controlling their effects. If the manipulations of DOC, DO, sulfide, pH, and temperature cannot explain the observed changes in the rates of methylmercury production and decomposition, dosing of the microcosms and mesocosms with environmentally relevant concentrations of iron would be a higher priority than selenium.

Comment: Cadmium is a well-known contaminant of phosphate fertilizers. Has cadmium been considered and/or measured in the Everglades and its biota. What are the relative levels of cadmium, and has a cadmium mass balance been attempted for the Everglades.

Response: Cadmium bioaccumulates to some extent in vegetation, but does not bioaccumulate substantially in higher aquatic animals. It does have a high affinity for organic matter and does accumulate in sediment. The District routinely monitors Cd in water and sediment at a number of sites in the Everglades canals and interior marshes. A review of the data indicates that Cd does not exceed its numerical Class III Florida Water Quality Criterion to protect aquatic life. There are no freshwater Sediment Quality Criteria for Cd. Sediments from a subset of these sites are being screened for toxicity using the Microtox (R) sediment bioassay and toxic sediments identified in this way will be tested for toxicity to fish and macroinvertebrates using standard bioassay methods adapted to the character of the sediments and conditions appropriate to south Florida. Should the sediments prove toxic, a Toxicity Identification Evaluation will be conducted. If Cd is present in toxic amounts, the bioassay and TIE should detect it. The cadmium concentrations at the District's major pump stations in the northern Everglades average 0.36, 0.23, 0.31, and 0.72 ug/L at S-5A, S-6, S-7, and S-8, while the average concentration in the interior marsh at WCA-3A-15 is 0.26 ug/L, indicating that there are neither high concentrations nor a significant spatial gradient suggesting substantial loadings from fertilizer contamination in the EAA.

Comment: What are the long-term effects of accumulations of sulfates from fertilizers? Sulfur influences the bioavailability of metals.

Response: Regarding the sulfur-mercury link, see pages 7-39 to 7-40 and 7-56 to 7-60 of Chapter 7 of last year's ECR (2000). Studies of other potential adverse effects of excess sulfate (e.g., sulfide toxicity to higher aquatic plants and animals) have not yet been conducted. The possible effects of agricultural sulfate in stormwater runoff are being investigated. Known and hypothesized effects of the sulfur cycle on the mercury cycle are summarized in Appendix 7-4. A presentation will be made at the Peer Review Workshop by the authors of Appendix 7-4.

Comment: It would be useful to have a table in the introductory material that showed the body burdens known to have caused problems in various mammals, birds (and humans), along with the mean (SE) and ranges for these species from the Everglades.

Response: As discussed above, our limited resources are devoted to areas where there is some potential for control. Many trace elements and organic contaminants may be affecting Everglades biota

Comment: The removal and retention of mercury in the STAs may be a function of the hydraulic retention time (HRT) as well as of the other variables. The hydraulic loading (HLR) and the nominal HRT (NHRT) are directly related, but there is little relationship between the actual HRT (AHRT) and the HLR as demonstrated in the study done on STA Cell 4. This needs to be clarified with respect to the mercury problem.

Response: HRT is believed to play a role in determining the removal efficiencies of Hg(II) and MeHg by STAs. See pages 7-42 to 7-44 and Appendix 7-5 of ECR (2000). The influence of flow and depth on inorganic mercury transport and disposition and methylmercury production and decomposition are taken into account explicitly in EMCM-II.

Comment: Could the deposition rate of mercury be considered when developing water quality criterion?

Response: The water quality criterion would be based only on biological effects. An attempt would be made to adjust the deposition rate downward to achieve the criterion. To the extent that some fraction of the deposition is non-abatable, that might be taken into account in quantification of the assimilative capacity of the ecosystem for the development of the TMDL mandated by the Federal Clean water Act, but not the water quality criterion.

Comment: The use of statistical differences needs to be clearer. Either something is statistically significant, or it isn't. "Not statistically definitive" is an odd way of putting it. The reasons should be stated explicitly.

Response: This was poor choice of wording. The figure speaks for itself; there are statistically significant declines in mercury in some biota over the last 3 years. This is certainly good news but we are uncertain as to its policy significance at this time. It would be premature to declare the problem solved without following this nascent trend further. Presuming this trend relates to previous declines in deposition, we are mindful of Likens' and Bormann's admonition, stemming from their work on acid deposition at Hubbard Brook, that atmospheric deposition trends are very noisy and that a 3-5 year record can be misleading.

Comment: Does massive development change the water flow - and what effect does this have on Hg?

Response: Yes. See pages 7-61 to 7-65 in ECR (2000). See also our response to Note 5.

Comment: Was Hg used in agriculture (as it was in many other places)? In New Jersey, historical use of mercury in agrochemicals (mainly as a fungicide) is believed to have contributed to ground water contamination.

Response: Some but not extensive agricultural use as a fungicide probably occurred in the 1930s through 1950s. There is no evidence of remaining use of inorganic mercury as a seed treatment or methylmercury as a turf fungicide today, and the vast majority of the EAA land is under sugar cane, which requires no inorganic or organic mercury pesticides for proper pest management. See also the next Note.

Comment: The assertion that 95 % of the mercury in the Everglades is derived from the atmosphere seems high, and the evidence needs to be put in one place. Further, this is stated as fact in some places (7-3), and as unclear in others. No doubt the original explanation was given in an earlier report.

Response: There is no evidence that any mercury other than that deposited from the atmosphere is affecting the Everglades. Stormwater runoff from the EAA (and presumably elsewhere) contains some mercury. Concentrations are consistent with the idea that the mercury in stormwater also originated from atmospheric deposition on the tributary watershed. Whether it comes from contemporary and potentially controllable deposition or from cultivation practices that are releasing mercury deposited earlier is unknown. STAs reduce concentrations of mercury in EAA stormwater. Rainfall falling on almost 4,000 square miles or  $1 \times 10^{10} \text{ m}^2$  to an average annual depth of 1.3 m averages about  $13 \text{ ug/m}^3$  THg (Guentzel, 1997) or roughly 170 Kg/yr, while dry deposition is expected to be anywhere from 50% to 100% of wet deposition. EAA runoff flows average about 800,000 acre-ft per year or about  $1 \times 10^9 \text{ m}^3/\text{yr}$  with an average concentration of about  $2\text{-}3 \text{ ug/m}^3$  or roughly 2-3 Kg/yr. Thus, the 95% figure is actually quite conservative. The evidence for the 95% figure is given and cited in earlier reports.

Comment: The drying, re-flooding, and effects of fire on MeHg in the Everglades systems needs to be more clearly noted in the first part of the report.

Response: Further investigations will be reported in ECR 2001.

## SPECIFIC COMMENTS ON CHAPTER 7

Comment: page 7-1 - the initial paragraph only talks about atmospheric deposition and waste incinerators, but there are other non-point and point sources (e.g. power plants), and these might be mentioned.

Response: They are mentioned elsewhere.

Comment: page 7-3 - The FDA does not analyze fish at the moment. - What are the advisories for humans eating fish? Has anyone worried about people eating the raccoons? From work elsewhere, I have found that some people eat raccoons (posing a potential Hg problem).

Response: The Health Department Advisories are on their web site and also on the DEP web site. A raccoon advisory has not been considered, but a criterion to protect the Florida panther must take into account its propensity to switch from preferred prey like hog and deer to less desirable prey like armadillo and raccoon when preferred prey are scarce

Comment: page 7-6. If the high Hg problem is in the summer, why was the study only conducted for one month? - should mention here how close the model approaches reality.

Response: SoFAMMS was a pilot study, albeit a most informative one, directed toward testing and validating field and laboratory methods and models for use in larger, longer-term studies. It was conducted during summer because approximately 85% of rainfall deposition of mercury occurs during that season.

Comment: page 7-7. I would like more information on the movement of mercury between MeHg and HgS (an insoluble sink). Under what conditions is mercury ever freed from HgS? - the overall problem of sulfate seems complex, and the report needs a section that deals specifically with its role in relation to the bacteria (and to other elements).

Response: Ravichdran et al. (1998) were able to demonstrate that Everglades DOC can redissolve HgS and prevent its precipitation under thermochemical conditions that would be calculated to require precipitation in the absence of DOC, but the implications of this effect for methylmercury production are not straightforward. This chemistry is under active investigation. Appendix 7-4 summarizes the known and hypothesized links between the sulfur and mercury cycles and the proposed research to test the leading hypotheses.

Comment: page 7-8. A clear definition of what would constitute evidence of harm to wading birds needs to be inserted.

Response: This will be done. The risk management goal is to protect, and where necessary restore stable nesting populations of wading birds that are reproducing successfully in the Everglades basin. Thus, any reduction in abundance or production of populations of piscivorous wading birds (decrease in population sustainability) resulting from methylmercury toxicity would constitute an unacceptable impact (i.e., harm).



Comment: page 7-13. What are the current methods used to test for bioavailable Hg (II)? This is a rapidly expanding field. Will there be in vitro or in vivo tests.

- Will the water samples be collected only from lakes in the Everglades, or from all of Florida?
- - Again, the question of cadmium is relevant to eutrophication from fertilizers. Has this been examined?

Response: This is an area of active research and presentations to the panel will address this question.

Natural waters from the Everglades and elsewhere will be examined. Other than the rate of methylation by SRB, there is no method now in use for estimating the bioavailability of Hg(II).. This rate may be affected by factors other than the bioavailability of Hg(II). However, recent developments in the area of genetically modified bacteria have made it possible to insert the gene that codes for the production of the bioluminescent enzyme, luciferase, after the mer operon that governs the production of the enzyme involved in heavy metal detoxification in E coli (Virta et al., 1995). The calibrated biosensor will make it possible to define functionally the fraction of Hg(II) that is bioavailable to and transportable across the membrane of the modified bacterium. See pages 7-12 and 7-13 of Chapter 7 on the project by Bill Landing and co-workers in Finland on this topic.

Other than routine District monitoring, there will be no cadmium process research studies.

Comment: page 7-14. Does the rule have a gradient that is different near the agricultural areas? or is the rule the same throughout the Everglades?

Response: How this rule is expressed and how compliance with this rule is to be determined are the subjects of ongoing debate that will be covered in the peer review of other chapters. See also response to comments from William Green on Chapter 3.

Comment: page 7-15 -what was the problem with the reproducibility of the results? A statement such as this needs explanation (even in the summary).

Response: These are preliminary results that will be more fully explored in the Phase II work. A more detailed analysis is not appropriate at this time.

These are preliminary results that will be more fully explored in the Phase III work. A more detailed analysis is not appropriate at this time.

Comment: page 7-16 - Do the models include other metals that alter Hg uptake or effects? Which are included or at least considered? - what are the uncertainties considered?

Response: The models do not consider the effects of other heavy metals like copper on Hg(II) complexation or selenium on bioaccumulation or toxicity or other metals like calcium and magnesium on MeHg biouptake. Uncertainties are taken into account by evaluating the sensitivity of model output to realistic ranges of input variables. When the Monte Carlo package is added to the EMCM-II platform, a probabilistic analysis of uncertainty can be carried out in a more rigorous fashion.

Comment: page 7-17 - It seems to me that a 50 % decline from local sources resulting in a 25 % reduction in deposition is substantial, and not one to ignore. Has this been modeled further? It would be useful to see the curve for declines versus reductions in mercury from local sources.

Response: The numbers cited were for illustrative purposes; were declines of that magnitude realized in practice, they would certainly be welcomed. Detailed description of the model uncertainties are given in the draft 'atmospheric modeling support document' for the TMDL Pilot Study (See Appendix 7-3). This and other TMDL documents a presently being re-drafted in response to a separate peer review and a final report will be issued this Fall.

Comment: page 7-19 - the rainy season is in the summer, yet the flights were in the winter - what is the effect of rain on the data collected?

Response: Additional flights were made in June, 2000.

Comment: page 7-20 - Neurobehavioral effects are left out of this discussion. Even subtle neurobehavioral effects might impair development and survival. - Why not use a species that is higher on the food chain (such as Great Egrets) that eat larger fish for the reproductive effects? Why not look at behavior of young Snowy Egrets? In other words, why not do both adult and young deficits on the same species? Also, why not look at Anhinga that also eat fairly large fish?

Response: Neurobehavioral effects are implicit in the studies of captive White Ibis and Snowy Egrets as well as in the study of fledgling survival of wild Great Egrets.

We have purposely focused on a few end points of concern to develop more in-depth, albeit narrower, information.

Comment: page 7-22 - Has the rule making occurred?

Response: The rule has not been adopted. A technical working group is developing recommendations. This will be followed by workshops to take public input. Then there will be rule adopting hearings

Comment: Page 7-23 - Will the TMDL be determined for the Everglades as a whole, or for parts?

- it would be useful to include a figure of the decline in mercury emissions in southern Florida.

- what is the level of the high hospital waste stream?

Response: There is no certainty about what will be done. The TMDL process was conceived to treat relatively straightforward water quality problems where there was a simple relationship between direct discharges and effects. Generalizing this concept to include the diffuse and indirect effects of air pollution sources presents a daunting technical challenge.

Estimates of emissions trends will be presented at the peer review meeting.

Hospital waste has been a persistently high mercury waste stream. EPA and DEP are approaching this as a pollution prevention problem. In 1999 EPA signed an agreement with the AHA to work toward eliminating mercury from medical products, equipment and processes.

Comment: page 7-28. What were the sample sizes for the graphs.

Response: When we began collections in 1989 we insisted on 12 samples per site; since the mid-90s FWC has collected ca. 20 to get tighter confidence intervals.

Comment: page 7-29 - what were the ranges, sample sizes? Why use nestling feathers rather than fledgling feathers (when they are fully formed and no longer have a blood supply

Response: The graph was reproduced from an abstract submitted by the authors and does not show the range, standard deviation, or sample size. This information will be added to the final report.

The District adopted the protocols used by Frederick et al (1997), because the data generated by these researchers is considered the background data for Everglades wading birds under the federal permit to construct and operate the STAs. However, the District agrees that, in principle, the collection of feathers from fully fledged juveniles could provide more useful, less variable information about integrated exposure than from chicks. The embryo, chick, and juvenile life stages are all considered of potential toxicological importance, but the sources, dispositions, and effects of methylmercury are quite different for each life stage. The chick may consume more per unit body weight per day in contaminated prey than a fully fledged juvenile, but the methylmercury uptake efficiency across the gut may be lower because the residence time of food in the gut is shorter. While the developing chick may be more sensitive to the toxic effects of methylmercury than the fully fledged juvenile bird, the growing feathers of the chick are an important route of methylmercury depuration, reducing its susceptibility to a toxic effect from methylmercury uptake. Conversely, the fully fledged juvenile is probably less sensitive to the toxic effects of methylmercury but more susceptible to it because the feather depuration pathway is foreclosed. This underscores the importance of combining well-designed pharmacodynamics and kinetics and toxicology studies. In principle, it would be preferable to use the feathers of fully fledged juveniles to gauge the integrated exposure of the organism to methylmercury over its entire post-hatching development, but fully fledged birds are much more challenging to handle than their younger, smaller counterparts when encountered on or in the vicinity of the nest and much more likely to leave the area if disturbed.

Comment: page 7-32 - why no mention of alligators in the first 30 pages?

Response: Alligators were discussed on pages 7-4 and 7-25 of ECR (2000). The results of a combined USGS-District study of juvenile alligators collected along a north-south transect throughout the Everglades were not available as of the close of the reporting period on April 30, 2000, but will be included in next year's report.

Comment: Other needs - may need to mention levels of Hg in eggs (although mentioned in appendices).

Response: This is covered in the appendices.

## COMMENTS AND QUESTIONS ON APPENDICES

Comment: A7-3-6 - I need more explanation for why the predictions would not be affected by uncertainties.

- what were the assumptions? - is there any evidence for demethylation in any of the fish? What is the evidence?

Response: Because these curves are approximately exponential, the time required for a percentage change is independent of the error in deposition rate. As stated, "The actual magnitude of the change in fish Hg is of course still dependent on the magnitude of the load reduction." Deposition from global background was assumed to be zero. Demethylation in fish is apparently very slow, as indicated by their high bioaccumulation factors and low Hg(II) concentrations in the liver and kidney.

Comment: A7-3-7 - Were predictions made for any species other than bass?

Response: Yes. The food web consists of detritus, periphyton, phytoplankton, zooplankton, bethos, shrimp, mosquitofish, , bluegill/warmouth/sunfish (grouped together), and largemouth bass.

Comment: A7-3-11 - what are the effects of other contaminants and water acidity on the models and processes?

Response: The biogeochemical processes affecting the net mercury methylation rate and its incorporation into the bases of the food web are under active investigation.

Comment: A7-3-12 - what tissues were used? - why not a composite or use older larger fish?

Response: Muscle tissue was used for largemouth bass. The data came from a study designed to assess risk to recreational fishermen. The three year old bass is the most common fish in creel census data. Mercury data for bass, including older fish, are converted to age three though an empirical correlation and then applied to the E-MCM.

Comment: A7-3-13 - I worry about the assumption that deposition is constant over the long run (since it is not)? - What is the effect of an assumption of equal deposition for each month, when you know it isn't? Also measured at different places, data not comparable.

Response: This is a pilot study now under peer review and subject to revision. Our immediate problem is to distinguish reactive gaseous mercury of local origin (sources near the Everglades) from background and to calibrate deposition rate. The source-receptor model will ultimately deal with the time variation of deposition rate and with spatial variation in deposition. For the discussion on this page, deposition was held constant so that the simulation could produce steady-state fish concentrations.

Comment: A7-3-26 - How realistic are the prey assumptions made here?

Response: The Everglades food web is a complex function of antecedent and current conditions of hydroperiod and nutrient availability. Information is limited. As with any model, we expect to calibrate to representative conditions and verify over a range of variables that offer some potential for management to alleviate mercury bioaccumulation.

Comment: A7-3-33 - Why so few for part of the curve?

Response: Not many 40 cm and longer largemouth bass were captured by electroshocking in this location.

Comment: A7-3-39 - What is the prediction of 50 % in 6-7 yrs based on?

Response: This includes the entire set of processes, assumptions and calibration data used in the E-MCM combined with simulation to steady-state over a long period of simulated time at constant deposition rate.

Comment: A7-3-45 - Why use Gambusia?

Response: There are many Gambusia data because they are ubiquitous in the Everglades and easy to catch. Their importance to higher trophic levels is uncertain.

Comment: A7-3-46 - What are the dampening effects that occur in three year old fish?

Response: Three year old fish integrate deposition rate and other processes that are changing faster than the three-year time increment and, thus, smooth out more rapid changes.

Comment: A7-3-50 What are the factors that control demethylation, and in what compartments?

Response: This chemistry will be discussed at the workshop. It is complex, but quite important in determining the net rate of production of methylmercury.

Comment: A7-4-4 - What are the latest data from the fire studies?

Response: See Appendix 7-8.

Comment: A7-5-5 - How were the cores dated?

Response: This question will be addressed by the presentation by Curt Pollman of Tetra Tech.

Comment: A7-5-12 - This and other reports within this chapter make it clear that a diagram of the currently assumed mass balance for the Everglades would be useful

Response: Agree. This will be added to the main chapter in a future report.

Comment: A7-5-19 Were the experiments described in task 3 done?

Response: These questions will be addressed by the presentation by Curt Pollman of Tetra Tech.

Comment: A7-8-1 - Was this difference due to fire? drawdown? both?

Response: Samples of water, sediment, vegetation, and fish were collected at 13 sites: three that burned and ten that didn't, and of the three that burned, only one was a routinely monitored site. The highest increase in methylmercury bioaccumulation in mosquitofish occurred at sites that dried out completely but did not burn. The reason for these differences are now under investigation.

Comment: A7-9-11 - There is sometimes great variation in Hg accumulation within the species within a genus of fish - was there no similar variation within Lepomis?

Response: Yes, there was; as outlined on page A7-9-41, sunfish species was found to be a significant factor in tissue mercury concentration (ANOVA on ln-transformed data,  $df = 3, 378$ ;  $F = 54.2, p < 0.001$ ), with mercury concentrations in *L. gulosus* (warmouth)  $> L. punctatus$  (spotted

sunfish) > *L. macrochirus* (bluegill) > *L. microlophus* (reardear sunfish) (Tukey test multiple comparison procedure,  $p < 0.01$ ). These interspecies differences in tissue mercury concentration were not a function of size differences; weight of *L. gulosus* = *L. microlophus* > *L. macrochirus* > *L. punctatus* (Dunn's multiple comparison procedure,  $p < 0.05$ ).

Comment: A7-9-14 - More detail needs to be given for the feather work. Which feathers were collected (primaries, body feathers) at what age? What age is a "nestling"? Hg levels vary at different ages of nestlings (that is why people normally take feathers after they are fully formed).

Response: First, it should be emphasized that, in accordance with USACOE permit 199404532 Condition 8b.2, results from the earlier study by Frederick et al. (1997) were found to be representative of background mercury concentrations in Everglades wading birds (FTN Associates, 1999). Accordingly, the District's monitoring program was designed to be consistent with that database generated by Frederick et al. Having said that, growing scapular feathers are collected from nestlings at 2-3 weeks of age. Nestlings at this age can still be captured without much difficulty.

Comment: A7-9-50 - What are the data for the regression of bill length to age?

Response: Regression of age on bill length was generated by Peter Frederick and Marilyn Spalding and remains unpublished.

Comment: A7-9-51 - The problems with use of feathers of nestling needs further clarification. Ages 12-23 are when chicks are growing rapidly, and mercury levels might be expected to vary greatly. If the regression was not significant, then maybe it should not be used. Further (next page), the regression may be a problem because older chicks are fed large foods (which should have higher mercury levels). For the yearly differences to be equivalent exactly the same-aged chicks (same degree of feather growth) would have to be compared? Was this done (Fig. A7-9-18).

Response: Standardized feather-THg concentration for nestlings (i.e., for a bird with a 7.1 cm bill) at JW1 colony (7.18 and 6.9  $\mu\text{g/g dw.}$ , **Table 7-9A-16**) did not differ between 1999 and 2000 (ANCOVA,  $df = 1, 20$ ;  $F = 3.04$ ,  $p = 0.19$ ). Because regressions of THg concentration on bill length was not significant for birds from L67 colony in either 1999 or 2000 (ANOVA:  $df = 1, 18$ ;  $F = 2.4$ ;  $p = 0.138$ ;  $df = 1, 8$ ;  $F = 1.7$ ;  $p = 0.229$ ; respectively), standardized concentrations were not calculated nor was ANCOVA used to assess between-year differences at this colony. Instead, between-year differences in feather-mercury concentration at L67 (i.e., arithmetic mean concentrations) were evaluated using a Student's t-test and were found not to be significant ( $t = -0.76$ ,  $df = 28$ ,  $p = 0.454$ ). This was a valid test because there was no between-year difference in bill length in nestlings sampled at L67 (means were 5.5 cm in 1999 and 5.3 in 2000;  $t = -0.494$ ,  $df = 24$ ,  $p = 0.625$ ). A similar t-test might have been used to compare feather concentrations in 1994 (mean age was estimated to be 16 days), 1999 (15 days old) and 2000 (16 days old); however, without seeing the actual age distributions, we felt uncomfortable performing the analysis.

**Table7-9A-16.** Standardized (least square mean for a chick with a 7.1 cm bill) and arithmetic mean concentrations of THg ( $\mu\text{g/g dw}$ ) in growing scapular feathers collected annually from of great egret nestlings at JW1 and L67 colonies.

Colony	LSM $\pm$ 95 <sup>th</sup> CI (mean $\pm$ 1SD, n)			
	1994 <sup>*†</sup>	1995 <sup>*</sup>	1999	2000
JW1	21.12 $\pm$ 6.1 (25.0 $\pm$ 7.9, 9)	14.51 $\pm$ 3.31 (NA, 8)	7.18 $\pm$ 1.14 (4.0 $\pm$ 2.2, 13)	6.9 $\pm$ 2.9 (3.4 $\pm$ 1.9, 10)
L67	16.29 $\pm$ 4.53 (NA, 27)	15.51 $\pm$ 6.16 (15.9 $\pm$ 6.16, 14)	NC (3.6 $\pm$ 1.5, 20)	NC (3.2 $\pm$ 1.4, 10)

\* Data from Frederick et al. (1997);  $\pm$  1 SD.

† Concentrations standardized to a bill length of 5.6 cm.

NC – not calculated where slope of regression was not significant ( $p > 0.05$ ).

Comment: A7-9-53 - How could they compare 21 day old chicks to those than ranged from 12-23 days of age? - the 1993 to 1999-2000 levels for eggs do not look significantly different - was this tested? Moreover 0.46 is very close to 0.50 (detrimental effects level listed). How was it concluded that there was no effect?

Response: As discussed above, the regression of feather-THg concentration on bill length (age) was not significant at the L67 colony and, thus, there is no reason to believe that levels would be substantially greater when the birds were 21 days old than the observed arithmetic mean. Based on that, we can directly compare the observed arithmetic mean, 3.2 mg/kg, with the 17 mg/kg in the dosed birds. Alternatively, at JW1, where the regression of feather-THg concentration on bill length (age) was significant, we calculated a least squares mean for nestlings with 7.1 cm bill (i.e., to be consistent with 1995 data - @ 24 days old: Frederick et al. 1997). This resulting value, 6.9  $\pm$  2.9 (LSM  $\pm$  95% CI) did not approach the feather-THg concentration of 17  $\mu\text{g/g}$  in the dosed birds when they were 21 days old. During the District's responses to comments during the October 5<sup>th</sup> panel, we plan to present additional data on the regressions that should make these comparisons more clear.

In 2000, egret eggs collected from JW1 and L67 colonies had a mean egg THg concentration of 0.37  $\pm$  0.16  $\mu\text{g/g}$  (fresh weight,  $n = 13$ ). This concentration was not significantly different from concentrations observed in 1999 (0.41  $\pm$  0.205  $\mu\text{g/g fw}$ ,  $n = 20$ ;  $t = 0.55$ ,  $df = 31$ ,  $p = 0.59$ ).

Based on a literature review, Thompson (1996, pg. 345) concluded that “mercury concentrations in eggs of up to approximately 0.5 mg/kg ( $\mu\text{g/g}$ ) appear to have little detrimental effect on reproduction.” Using the mean egg-THg concentration observed in 2000 as a guide (i.e., 0.37), it appears that the risk of adverse effects from current in ova mercury exposures to the majority of the egret population is low.

Comment: A7-10-1 - Have you looked into cadmium? - Why not reduce amount of water lettuce mechanically (removing Hg from the system)?

Response: The focus of this study was ultra-trace sampling and analysis for total mercury and methylmercury. Mechanical harvesting of floating macrophytes or phytoplankton mats has been considered as a phosphorus removal mechanism, but the cost of collection, dewatering, and transport for processing have been calculated to be disproportionate to the removal efficiency.

Comment: A7-10-8 - What was the vegetation composition and habitat above and below this area? Did the methylation rates vary?

Response: At the northern test site, the vegetation above and below is monotypic cattail with some open slough area, while at the southern site, the vegetation is that of a mixed marsh community with cattail predominating, again with some open slough area. This was a straightforward mass budget study. No methylation rates were measured or can be inferred because of the non-detectable concentrations in the outflow waters.

Comment: A7-11-1 - Why not conduct it for the whole year, thereby having information for summer when methylation rates are highest?

Response: Mosquitofish were collected quarterly from September 1997 through February 2000. Only the choice of sites changed.

Comment: A7-11-4. I need more information on pH and its effect on total mercury loading and methylation rates?

Response: The formula developed by Hakanson (1980) requires the input of the concentration of water column TP, the concentration of THg in the sediment, and the water column pH for the calculation of the concentration of MeHg in a 1-Kg northern pike or equivalent. Hakanson (1980) obtained this empirical relationship between fish THg and THg in sediment, pH, and BPI through correlation analysis of published data from primarily Scandanavian lakes. However, there is a mechanistic rationale for the influence of each of these variables in Hakanson's formula. BPI is a surrogate for the physical and chemical manifestations of trophic state, including organic particle production, settling, decomposition, and dissolved oxygen consumption, and for the biological manifestations of trophic state, including the relative proportions of saprotrophs and autotrophs in general and food web structure in particular. Low pH (high acidity) is believed to increase the lability of complexed Hg(II) in the water column and sediment pore water, facilitating methylmercury production and bioaccumulation. In addition to the rate of MeHg production being proportional to it, the concentration of THg in the sediment is a surrogate for the net THg flux to the system, because the net THg flux divided by the rate of sediment accumulation is the THg concentration in the sediment. The empirical formula developed by Exponent (1998) incorporates only the influence of water column TP and omits that of sediment THg and water column pH determined to be important by Hakanson (1980). However, while the Hg(II) in the sediment increases by about 50% between F1 and U3, the pH does not change substantially.

Comment: A7-11-16 - What is the evidence for more complex food webs (top of page)?

Response: The mosquitofish collected from each site are in the same size range. The evidence that another step has been added in the food chain comes from unpublished gut content studies by Paul Garrison of the Wisconsin DNR, which demonstrate that mosquitofish at F1 have more benthic infauna and detritus in their guts than the mosquitofish collected at U3. Unpublished District data indicate that the ratio of MeHg to THg increases monotonically from less than 25%



at F1 to greater than 85% at U3, suggesting that U3 mosquitofish are feeding at least one step up on the aquatic food chain from the F1 mosquitofish. This latter deduction is based on the published observation that MeHg is much more efficiently absorbed than Hg(II) across the gut membrane of fish but that Hg(II) is present in detritus along the WCA-2A "F" transect in concentrations on the order of 100 times greater than MeHg. Corresponding data for benthic detritivores are unavailable. However, some of this apparent difference may be an artifact of the analysis of the mosquitofish without purging their guts of detritus and undigested prey, but scoping calculations suggest that this is only likely to be a first-order correction for F1, where mosquitofish average about 10 ng/g wet wt, so it is likely that the mosquitofish have absorbed both Hg(II) and MeHg from the detritus and detritivores, albeit with different absorption efficiencies.

Comment: A7-13-2 - Here and elsewhere, information on pooling is seldom given (how many fish, or what size, why pool).

Response: Mosquitofish (*Gambusia holbrooki*) is the only species that is pooled. Sunfish, largemouth bass, bird eggs, bird feathers are all analyzed on individual basis. The District's Standard Operating Procedure calls for the collection 100 -250 individuals from all size classes (small = <0.07 g, medium 0.07-0.28 g, and large >0.28 g),

Pooling, i.e., composite sampling, can increase sensitivity, reduce intersample variance effects, and dramatically reduce analytical costs. Composite sampling increases the amount of material available for analysis and, hence increases sensitivity to achieve lower method detection limits. This is particularly important for small-size mosquitofishes (= small mass). Increasing the amount of available material also is important for use as quality control samples, e.g., splits between labs, duplicate digest, matrix spikes, sample archiving. Cost effectiveness is always an important consideration in any large-scale routine monitoring program.

The District carried out a study where individual fish and different pooled samples were analyzed. At that time, we estimated a minimum of 0.256 grams of sample (at a tissue concentration of 5 ng/g or 1.28 grams at 1 ng/g), i.e., aliquot to be digested, would be required to archive the method detection limits (0.13 to 0.42 ng/vial). At the same time, based on the observed variance in tissue mercury concentration among medium-size individuals, a minimum sample size of twenty individuals ( $n = 20$ ) would be needed to statistically detect a 30% difference between site means of 0.05. This study was conducted at the ENR Project, where concentrations were relatively low compared to the rest of the Everglades and, thus, represent the smallest signal to noise ratio. For these reasons, the District chose to composite mosquitofish.

Comment: A7-13-7 - why are there only 6 points, and what are the dots on the next graphs?

Response: The data points (and calculated BAFs) represent only sites where water and mosquitofish were co-sampled. Only six water samples were collected.

The filled circles in **Figure 7-13A-3** represent data that lie outside the 10<sup>th</sup> and 90<sup>th</sup> percentile (i.e., whiskers of the boxplot). The caption will be revised in the final draft to reflect this.

Comment: A7-13-13 - Are there plans to study movement of bass and other key fish? Movement of fish is mentioned but it's not clear that it will be examined so the impact on the spatial data can be determined.

Response: There had been some speculation that sampled largemouth bass populations may not be representative of STA 6 conditions. Specifically, it was theorized that bass could move

large distances and confound spatial interpretations. Regrettably, due to budget constraints and reallocation of resources, a study of fish movement is not being considered at this time. However, the District is moving forward with adding sunfish to the STA monitoring program. Because sunfish are thought to have smaller ranges, to a degree, this may reduce spatial uncertainty.

Comment: A7-13-14 - I would like to see the data on translating from fillet to whole body burden. What fish was this done for? How many individuals were involved? Was it on the basis of wet or dry weight?

Response: Largemouth bass (*Micropterus salmoides*) is the only fish species that the District is currently analyzing as fillets. Sunfish are analyzed as whole-fish homogenates. The regression reported by Lange et al. (1998; cited in appendix) was for largemouth bass (on a wet weight basis). He did not report degrees of freedom, but I count at least 27 data points in his graph ( $Y_{int} = -0.0057$ , slope = 0.6945;  $r^2 = 0.9915$ ). These results were remarkably similar to the relationship reported by Bevelheimer et al. (1997; whole-body concentration in mg/kg =  $0.7 \times$  fillet concentration in mg/kg).

Bevelheimer, M.S., J.J. Beauchamp, B.E. Sample, and G.R. Southworth. 1997. Estimation of whole-fish contamination concentrations from fish fillet data. Prepared by the Risk Assessment Program, Oak Ridge National Laboratory, Oak Ridge, Tennessee 37831. Prepared for the U.S. Department of Energy, Office of Environmental Management.

Comment: A7-14-7 - If I understand correctly, they were sampled months after the start of the experiment. Would the levels have been higher immediately after? This is especially true if there were heavy rains prior to your sampling.

Response: Yes. However, mosquitofish sampling was only added to the start-up study after it became apparent that water column MeHg concentrations in Cell 5 had increased 100-fold over those in adjacent Cell 4.

Comment: A7-14-10 - While biodilution does occur, once most of the growth occurs, then the Hg levels will increase because of continued exposure. What is actually lost to the system? and how?

- although the fish from cell 5 were small compared to S-5a, why not compare only those that were of equal sizes? - what about selenium levels here?

Response: There is a distinction between growth dilution and plant-mediated biodilution. There were insufficient numbers of fish in Cell 5 to allow subsampling by size cohort viz S-5A. The focus of the study was on the bioaccumulation of THg as MeHg in mosquitofish, sunfish, and largemouth bass in response to the pulse of MeHg production following flooding. Selenium may mediate bioaccumulation and toxicity, but monitoring routinely for it in water, fish, vegetation, and sediment would have more rapidly depleted already limited resources than could be justified based on the likely magnitude of its influences and the likely differences in its concentrations across the Everglades.

Comment: A7-16-3 - here they speak of juvenile egret feathers. As mentioned above, exactly what are these? size, age?

Response: Should be pre-juvenile nestling feathers. Error will be corrected in final draft.

Comment: A7-16-5 - I find this table difficult to read and interpret. What is being shown? Sample sizes?

Response: Mean (S.D.) and Range of THg residues in largemouth bass flesh and nestling great egret feathers. Sample size is indicated in column labeled "N". Typographical errors were introduced in electronic transmission in the date ranges of sample collection.

Comment: A7-16-7 - It seems that fish have been collected from the Everglades for some years. Why not do a sensitivity analysis so that you know how many fish you need to collect to see if there is a statistical difference. - I wouldn't say there was a decline in mercury if it wasn't tested because of small sample sizes.

Response: The need to consider largemouth bass age as a covariate in an analysis of covariance (ANCOVA) complicates the power analysis. Further, use of the ANCOVA itself is complicated due to the assumptions, including (1) that regressions are simple linear functions, (2) that regressions are statistically significant (i.e., non-zero slopes), (3) that the covariate is a random fixed variable, (4) that both the dependent variable and residuals are independent and normally distributed, and (5) that slopes of regressions are homogeneous (parallel).

After performing this statistical method over the last few years, Lange et al. came up with a target sample size of 20 fish from each site. However, there is no guarantee that 20 fish can be collected at each site, nor that the sampled fish will have an adequate age distribution for performing the regression, nor that all the other assumptions for ANCOVA will be met.

With regard to STA 6, as reported in A7-9, largemouth bass collected during both annual collections at STA 6 showed higher tissue mercury concentrations at the outflow as compared to inflow (**Table 7-9A-5**). While this difference between inflow and outflow was shown by ANCOVA to be significant in 1998 ( $df = 1, 26$ ;  $F = 22.9$ ,  $p < 0.0001$ ), because of an interaction between the effects of fish age and location on mercury concentration, ANCOVA could not be used to statistically evaluate spatial differences in 1999 (i.e., slopes were not parallel;  $df = 1, 35$ ;  $F = 4.65$ ;  $p = 0.04$ ). Notwithstanding the statistical limitations, bass collected at the outflow of STA 6 in 1999 clearly had substantially greater concentration of mercury than bass collected at the inflow. Notice that, similar to the mosquitofish, the degree of difference between inflow and outflow lessened in 1999 when levels of mercury in outflow fish decreased. The decline in mercury concentration in outflow bass from 1998 to 1999 was not significant, however (ANCOVA two-tailed test;  $df = 1, 36$ ;  $F = 3.73$ ,  $p = 0.12$ ).

Levels of mercury in bass from the interior of STA 6 must be interpreted carefully for a number of reasons. First, bass were collected from Cell 3 in 1998. In 1999, an effort was made to sample bass from both Cell 3 and Cell 5; however, only three bass were collected from Cell 5. The small sample size in 1999 and the lack of a significant age regression in 1998 (i.e., mercury concentration against bass age;  $df = 1, 16$ ;  $F = 0.0146$ ;  $p = 0.9$ ) did not allow for a valid test of between-cell differences on age-standardized basis. Nonetheless, mercury appeared to be at higher levels in interior bass from Cell 3 in 1998 compared to bass at the outflow, as well as interior fish collected from Cell 5 in 1999.

Comment: A7-16-8 - Do you have more information on the young of the year bass that had higher levels? Did they continue to increase? Is this a problem that will increase with changes in water levels and re-flooding (with fires)?

Response: The results from the fall 2000 collection are not available as of this writing.

Comment: A7-16-9 - what are the conservatisms built in? -were tissue levels of great egret chicks examined (some usually die from inadequate food, and these could be analyzed)? -it is very difficult to find sublethal effects in whole populations unless you know the levels in

individuals.- I would appreciate some maps of the relevant colonies, the foraging distances, the evidence that the adults do not feed near the colony in the critical egg-production phase.

Response: The conservatisms built into EPA's guidance level stem from their use of the mallard-LOAEL to establish their reference dose. In October 6-7, 1999, FDEP, SFWMD and University of Florida sponsored a workshop to address effects assessment for methylmercury in fish-eating birds (River Ranch, Florida). One conclusion that emerged from that meeting was that we should discontinue reliance on the mallard, which is grainivorous, for setting guidelines for piscivorous birds. To this end, this District initiated a collaborative project (obtaining the necessary permits, etc) with Gary Heniz, which is currently working on the CALFED project, to dose wading bird eggs. Unfortunately, because of budgetary constraints and reallocation of resources, this study has been terminated.

Response: While the District obtained federal and state collecting permits to salvage dead birds to attempt to do exactly what you suggest, we have not come upon fresh dead birds. The District attempts to take a minimalistic approach to sampling to conserve man hours and to reduce stress to the birds. Colonies are visited only 2-3 times a year. Consequently, while a few dead nestlings have been found, their condition did not warrant salvage.

Regarding our ability to find sublethal effects, we stated in Appendix 7-3b (ECR, 2000) that "we may simply be unable to detect population-level effects at our current level of effort." Appendix 7-3b further states that "although population-level effects have not been demonstrated, inferential evidence of negative effects to the individual continues to mount (Spalding et al., 1994; Sundlof et al., 1994; Beyer et al., 1997; Frederick et al., 1997, Bouton et al. 1999). Individual-level effects should not be dismissed out of hand, particularly in regards to the endangered Wood Stork."

Maps of the colonies with foraging ranges will be made available at the District's presentation to the panel on October 5<sup>th</sup>.

Regarding the statement about adults not feeding near the colony during the critical egg-production phase, as reviewed in Appendix A7-9 (ECR, 2001 draft), because albumen-mercury is strongly linked to dietary mercury (Walsh, 1990), levels in eggs appear to reflect exposure over a comparatively short period of time, possibly one or two weeks prior to egg laying (Fossi et al., 1984, Furness, 1993). Therefore, depending on the timing of the bird's arrival on the nesting grounds, mercury concentrations in eggs can closely reflect local contamination. Appendix 7.3b (ECR 2000) contains a lengthy discussion of scaling issues, which are critical in risk assessments. This discussion raises, among other things, the possibility that the female may integrate her exposure over a larger area prior to egg laying (i.e. relative to an adult feeding a nestling and, thus, the nestling, itself). This was based on two simple suppositions. First, prior to nesting (e.g., prior to roosting in future nesting colony, pair formation, establishment of nest territory, etc), individual birds may be wider-ranging within the basin (or sub-basin) compared to a bird feeding a nestling. This supposition was based on data from the USACOE systematic reconnaissance flights that showed large numbers of birds moving within the basin just prior to nesting (for discussion of SRF and citation see Appendix A7-9). For example, in February 1999, 12,720 birds were foraging in WCA2A. Numbers of birds decreased rapidly with only 1,000 birds remaining in May. At the same time in February, very few birds were observed in vicinity to L67 or known colonies in southern WCA3A. The second supposition is that prior to supporting nestlings, adult birds roosting at the nest colony would also likely have larger [more expansive] foraging range. Alternatively, while feeding a nestling the adult bird may forage in a relative small area. Although, not discussed in Appendix 7.3b, this is particularly important in the Everglades where the drying front (typically occurs during the period when nestling are being fed) concentrates

large numbers of birds in small areas. As such, a nestling might be considered more vulnerable than the egg, because exposure is integrated over a smaller area (i.e., that might contain a “hot spot”).

## **APPENDIX 7-3B: METHYLMERCURY RISK TO EVERGLADES WADING BIRDS**

### **OVERALL.**

This is overall an excellent study.

Comment: Were the levels of other contaminants that affect uptake and effects measured or considered (such as selenium)?

Response: The District does not routinely monitor constituents in eggs or feathers other than mercury. The available data for selenium (Rumbold et al. 1997; Frederick et al., 1997; P. Winger, USFWS, pers. comm) and cadmium (Spalding et al., 1997, P. Winger, USFWS, pers. comm) is insufficient to determine if interaction is currently affecting mercury uptake or effects. However, if the need for such data becomes important, the District does have archived material of both eggs and feathers for possible reanalysis.

Comment: One of the things that is needed to validate these and other models with waterbirds is to collect some data on feather levels compared to internal tissue levels (kidney, liver) from different aged chicks under experimental conditions. As a start, this could be done from egrets that died in the wild (there are surely some each year of different ages).

Response: A lot of these data are available in Frederick et al. (1997), but they have not been fully analyzed, yet. With regard to salvage of dead birds, please see previous response.

### **SPECIFIC**

Comment: page A7-3b-2 - I am slightly worried about not exposing the young birds from a day or two (Especially since in some of our work we find exposure at day 2 has a much greater effect than at age 10 or 12 days). If I read this correctly, dosing was done at 15 days of age (after much neurodevelopment had already taken place). An important next step would be to dose birds at an earlier age.

Response: Comment will be passed along to Peter Frederick of the University of Florida-Gainesville.

Comment: page A7-3n-4 -At the bottom of the page it says chicks were dosed from 20-98 days; how does this relate to the dosing at 15 day? Further, the toxicodynamics are not well worked out, and lab studies of levels in kidney and liver would be beneficial.

Response: Comment will be passed along to Peter Frederick of the University of Florida-Gainesville.

Comment: page A7-3n-4 - I found it hard to understand the different studies and the sample sizes. These may have been presented in separate reports, but it would have been helpful to have a summary table of the different studies.

If the risk assessment is reiterated, a table will be included that summarizes the various effects studies.

Comment: page A7-3b-12 : It is certainly true that more laboratory and field studies with these species are needed.

Response: DEP agrees that such studies would reduce the uncertainties in the extrapolation of the mallard duck NOAEL to other bird species and the corresponding need to use adjustment factors in calculating toxicity reference values for ecological risk assessments and water quality criteria for State Water Quality Standards to protect fish-eating birds.

Comment: page A7-3b-15: are there results from after the fire when Hg levels were much higher in the fish?

Response: No, they were collected after the April 30<sup>th</sup> deadline and, thus, will be reported in next years report.

Comment: page A7-3b-21 -top, what was the variance and range?

Response: In 1999, egg-THg concentration was  $0.41 \pm 0.205 \mu\text{g/g fw}$ .

Comment: -bottom - I would be careful about drawing conclusions back to the post-ecp conditions. The question really is, are there effects in the hot spots (especially if some of the larger colonies are nearby).

Response: While exposure models can be useful, some degree of model verification is important to reduce uncertainty. The easiest way to do this is to go into the field and look at the birds themselves. Unfortunately, wading birds do not regularly nest in WCA-2A. In 1998, three small colonies were located in WCA-2A. In 1999, a single mixed species colony of less than 100 nests was recorded in WCA-2A. In 2000, there were no colonies greater than 40 nests in WCA-2A (Gawlik, 2000). Consequently, the District monitors mercury in two great egret colonies in WCA-3A that have relative long-histories in the same location (i.e., JW1 and L67, which were survey by Frederick et al. in 1994 and 1995). After exposure in the central basin was modeled, resulting risk estimates (e.g., dose rates, HQ, predicted tissue concentrations) could then be compared to real world conditions (e.g., observed levels in eggs and feathers, reproductive success), ultimately providing perspective to the risk estimates (from the same model) for WCA-2A.

Comment: page A7-3b-22 -any data from 2000?

Response: The probabilistic exposure assessment was not reiterated in 2000 because of small sample size. At individual sites where sample size can become limiting, data from multiple years was used (i.e., assessment at U3 and 3A-15 was based on 1997-early1999). Further, because drydown was not caused by ECP, it was not related to the explicit risk hypothesis regarding TP.

Comment: page A7-3b- While I agree that estimating reproductive success only to 14 days is a bad idea, it is important to assess the effect of mercury exposure on eggs and chicks up to 14 days of age (when critical neurobehavioral deficits occur).

Response: We will pass this comment along to Peter Frederick of the University of Florida at Gainesville.

Comment: page A7-3b- - if a some or a lot of the population isn't nesting, could this be due in part to elevated mercury levels interfering with either behavior, feeding ability, or endocrine functions?

Response: Frederick et al. (1999a) have also suggested this possibility. While intuitively sensible, available evidence does not support this theory. The three-year running average for great egret nests in the Everglades have increased since the mid-1980s, with 1999 nesting estimates possibly exceeding nesting in 1940s (Ogden, 1999). Based on nest surveys and systematic reconnaissance flights estimating the abundance of wading birds in the WCAs, Gawlik (1999) estimates 87 percent (1998) to 89 percent (1999) of the waders (all species) nested in the last two years. Nesting in south Florida increased by 40% in 2000 to 39,480 nests. Best year since 1940s for ibises, since 1970s for wood storks (Gawlik, 2000). ”

Comment: - It is interesting that the breeding Great Blue Herons had higher Hg levels in feathers than non-breeding birds. It would be helpful to know the timing of the feather sampling with respect to the breeding and molt cycles. Is it known where these birds range when they are not breeding (e.g., in or out of the hot spot areas). If feathers were collected at the beginning of the breeding season, then levels reflect exposure largely during the non-breeding period (and differences may be difficult to interpret).

Response: Breeding status and reproductive success may be more strongly correlated with overall vigor of the males and females at the time of pairing, as reflected by foraging efficiency, rather than with the MeHg residues in the brain or reproductive organs. If this were the case, then the one might expect higher average concentrations of THg as MeHg in feathers in breeding pairs than in non-breeding birds.

Comment: page A7-3b-25 (top), finding consequences is difficult since you cannot examine all birds in the wild, and thus may not examine (by chance) those that are at the high end of exposure.

-the losses due to contaminants such as mercury may be offset by other causes of mortality and deficits. Effects are much better determined in laboratory conditions, especially given your level of exposure from mercury in the environment. Where are the "clean" birds in your Everglades colony to compare them to?

Response: At present we do not have a clean colony, FDEP has contracted Peter Frederick to find a clean colony. For their nestling dosing study, Frederick et al. collected eggs from a colony (identified as Alley North) located in northern WCA3A near WCA2A.

Comment: - It would be useful to know more about the Wood Storks. Are there no dead eggs and young that can be analyzed?

Response: While the District obtained federal and state collecting permits to salvage dead birds or addled eggs, this does not include endangered species.

Comment: - the conclusions (no. 1) is important, and suggests work needs to be done on Great Blue Herons.

- isn't one of the major colonies situated in the hot spot?

Response: The District's assessment concludes there is reason for concern, particularly regarding effects to the individual. Specifically, Conclusion 7 states "methylmercury risks to wading birds feeding exclusively in the WCA3A methylmercury "hot spot" (i.e., WCA-3A-15) are of potential concern and warrant further studies; some of which are already underway. However, this area is minimally affected by EAA discharges at present and this is not expected to change as a result of the ECP"

Comment: page A7-3b-26 : how does conclusion 5 follow from number 1?

Response: Conclusion 1 refers to risk to the wading bird if exposure were integrated over the entire Everglades basin. Higher exposures and elevated hazard quotients were forecasted when only fish from central Everglades sites were used. As evident from Figure 7, exposures and risk at WCA-2A-U3 were much lower, compared to the entire Everglades basin, the central Everglades basin and compared WCA-3A-15.

This risk assessment only attempts to test the hypothesis that risk to wildlife from MeHg will increase to unacceptable levels because phosphorus removal will reduce MeHg biodilution in the Everglades marsh downstream of the STAs (also known as the “inverse relationship effect”). WCA-2A-U3 was selected to represent the Post-ECP reference site because TP concentrations in the water column are typically below the 10 ppb criterion adopted in the Act as the default threshold. Because it is minimally impacted by nutrients, U3 has served as a reference site in earlier studies of the Everglades, and represents baseline conditions toward which restoration will be directed.



## Chapter 7: The Everglades Mercury Problem

### Responses to Questions Asked Prior to the October 6, Peer Review Panel Workshop by Dr. Joseph V. DiPinto.

By Thomas Atkeson & Paul Parks (DEP) and Larry Fink  
and Darren Rumbold (SFWMD)

Comments in this section are from Dr. DiPinto

Comment: I would like to hear more detail on planned next phase mesocosm studies to sort out the relationship of S, nutrients, and Hg loading on MeHg production and bioaccumulation, especially the stable Hg isotope experiments.

Response: This will be presented by Dr. Krabbenhoft and Dr. Gilmour at the October 5, 2000, workshop.

Comment: What has been learned since the 1999 report on P-MeHg bioaccumulation relationship? This is related to the adequacy of the 12 ng/L Class III WQS relative to fish levels/advisories. Currently, data-based relationship is weak.

Response: See pages 7-37 to 7-39 and 7-54 to 7-55 of ECR (2000) and Appendix 7-11 of draft ECR (2001).

Comment: The E-MCM model is very sensitive to K<sub>pl</sub> for MeHg. What is the basis for saying K<sub>P</sub> is not a function of algal species composition or physiological state? Is K<sub>pl</sub> different in WCA-1 vs WCA-2? Is K<sub>pl</sub> different along transect (gradient of P) in WCAs?

Response: This will be addressed by Dr. Pollman at the October 5, 2000, workshop.

Comment: Page 16 briefly notes modifications to E-MCM that will allow linkages w P models, incorporate S dynamics, allow a “bottom-up” bioenergetics representation. Can you explain what these are in more detail and will they assist in addressing the P, S, and Hg interaction issues?

Response: This will be addressed by Dr. Pollman at the October 5, 2000, workshop.

Comment: Significant local Hg emission reductions have been achieved, but their effects have not been seen in deposition monitoring. Why is this? Is it a monitoring problem or is it a real result?

Response: This is likely to be a consequence of beginning rainfall monitoring in 1994, after most of the emission reductions, which occurred in the period 1987-1993. Work is in progress on sediment core analysis to determine deposition changes.

Comment: Statement on p. 24 says that STAs sequester incoming Hg rather than enhance its release. Does this finding refer to THg or just MeHg? What is the magnitude of the impact of hydroperiod on this finding? Could be very important for design and operation of STAs.

Response: STAs sequester both THg and MeHg. See Appendix 7-5 of ECR (2000). The proportion of flow to rainfall, water depth, and HRT do affect THg and MeHg removal efficiencies. Perhaps surprisingly, higher flows and deeper water remove more THg and MeHg by decreasing the relative contribution of Hg(II) from rainfall and increasing contact cross-sectional area with plants. See Executive Summary of Chapter 7 (New Findings), pages 7-42 to 7-44 of ECR (2000), and Appendix 7-5 of ECR (2000).

## CHAPTER 7: THE EVERGLADES MERCURY PROBLEM

### Responses to Questions Asked Prior to the October 6, Peer Review Panel Workshop by Dr. Donald Kent

By: Thomas Atkeson & Paul Parks (DEP) and Larry Fink  
and Darren Rumbold (SFWMD)

Comment: Does the SFMSP include industry representatives?

Response: Yes, the utility industry, as represented by EPRI, the Florida Electric Power Coordinating Group, and Florida Power and Light, has supported substantial contributions to the work of the SFMSP. The Sugar Cane Growers Cooperative of Florida has participated in SFMSP meetings and provided risk assessment analyses of mercury risk to wildlife through its consultants, PTI (now E<sup>x</sup>ponent). All meetings in which DEP participates are public; no one is excluded.

Comment: At what rate do organisms eliminate MeHg? Is this considered in the ecological risk model(s)?

Response: Depends on species, body weight, and reproductive status, but, in general the half-life increases exponentially with body weight. For example, a mosquitofish has a half-life on the order of days, which explains its rapid response to changing environmental conditions, while the half-life in an age class 3-yr largemouth bass is on the order of several years, which explains why it is an excellent integrator of environmental conditions. In addition to excreting Hg more efficiently than fish, birds and mammals eliminate methylmercury in growing feathers and hair. The half-life of methylmercury in humans is approximately 45 days, as determined by radiolabeled MMHg dosing studies.

The District maximized the use of empirical data and minimized the use of model estimates in performing its deterministic ecological risk assessment (DERA) (See Appendix 7-2 in ECR, 1999) and probabilistic ecological risk assessment (PERA) (See Appendix 7-3b in ECR, 2000) for post-ECP MeHg exposures to fish-eating animals. This was achieved by adopting a reference site approach. The representative “already impacted area” chosen for this exercise was that downstream of the S-10 structures in WCA-2A along the “F” research transect. The well studied oligotrophic site, WCA-2A-U3, was chosen as representative of unimpacted phosphorus and mercury conditions. To implement the reference site approach it was necessary to assume that, if the most eutrophic of the well-studied sites along the “F” transect, F1, is restored to U3-like oligotrophic conditions, then the post-restoration THg concentrations in mosquitofish, sunfish, and largemouth bass at F1 will become like they are now at U3. The adoption of U3 as the reference site allowed the District to use measured rather than modeled THg concentrations in small, medium, and large fish collected at pre-restoration U3 in 1997 through early 1999 as a surrogate for post-restoration F1. Based on this approach, the District concluded that there would

be some, but not an ecologically significant, increase in post-ECP methylmercury risks to fish-eating wildlife in the restored areas of the northern Everglades.

A reduction in the average annual mercury loading rate to the Everglades is likely to reduce the average annual net methylmercury production rate with a corresponding decrease in the concentrations in small, medium, and large fish over time, with lag times on the order of weeks, months, and years, respectively. The Everglades Mercury Cycling Model-II, which is being developed for the Department and the District by Tetra Tech, Inc., can account for the effect of a reduction in inorganic mercury load on net methylmercury production and the effect of differences in methylmercury elimination rates in small, medium, and large fish on the methylmercury concentration distributions in those populations as a function of fish age and time, which would mitigate the post-ECP risk estimates based on a constant methylmercury concentration in fish. However, the District assumed a constant concentration of total mercury as methylmercury in fish over time for purposes of quantifying the post-ECP risks to fish eating wildlife, so its conclusions are not dependent on decreasing methylmercury concentrations in fish over time.

Comment: Does the 1 to 5 percent of mercury entering the Everglades from watershed runoff merit management? For example, does mercury in watershed runoff, added to mercury deposited from the atmosphere, cause total mercury levels to exceed critical thresholds? Conversely, if mercury in watershed runoff is eliminated, will ecological risk decrease significantly? If atmospheric mercury is the sole determinant of MeHg levels in the Everglades, then our efforts might best be directed at reducing local and regional sources, and determining if the STAs or ATT exacerbate MeHg.

Response: Stormwater runoff from the EAA (and presumably elsewhere) contains some inorganic mercury and methylmercury. Whether it comes from contemporary and potentially controllable deposition or from cultivation practices that are releasing mercury deposited earlier is unknown. The inorganic mercury and methylmercury in the northern primary canals probably originates almost exclusively with EAA runoff and Lake Okeechobee releases. The average concentration of total mercury in canal water entering WCA-2A through the S-10 structures is about one-half and one-third the corresponding average concentrations at the most eutrophic and the most oligotrophic of the well-studied sites along the nutrient gradient and almost an order of magnitude lower than that in rainfall, so there may be some localized dilution of the atmospheric deposition load in the immediate downstream vicinity of the S-10 structures. For those STAs that perform like the demonstration-scale STA (ENR Project), the inorganic mercury and methylmercury in EAA runoff and Lake Okeechobee releases treated by those STAs will be decreased, not increased. This will increase the magnitude and extent of the beneficial effect of the dilution afforded by the S-10 flows in WCA-2A. However, the area influenced by this localized beneficial effect is dwarfed by the area that is almost solely under the influence of atmospheric deposition. This flow dilution phenomenon is discussed on pages 7-61 and 7-62 of ECR (2000). Thus, as you suggest, the restoration strategy is directed towards control of atmospheric sources with potential control of methylation rate as a supplementary strategy. Both DEP at the state level and USEPA nationally have devoted considerable effort to eliminating the commercial and industrial uses of mercury and controlling significant emissions sources. Incinerator emissions in southern Florida, for example, have decreased > 90% since the mid-1980's, based on annual stack sampling pursuant to CAA permit requirements.

Comment: At what depth does Hg accumulate in the soil? At what depth is Hg sequestered and unavailable for conversion to MeHg?

Response: Dated cores of Everglades wetland soils show the accumulation rate of mercury increased approximately 5-fold between 1900 and 1990. Inorganic mercury and methylmercury sorbed to settling undecomposed plant biomass are generally considered to be in one of two forms or pools: loosely bound or labile and strongly bound or refractory. Some exchange occurs between these two pools, mediated by various physical, chemical and biological processes and factors. However, the availability of inorganic mercury for methylation and other reactions such as those leading to evasion back to the atmosphere is uncertain. Nevertheless, one can answer this question in broad terms by some simple scoping calculations.

The average peat soil deposition rate has been calculated to be in the range of 0.1 to 1 cm/yr depending on location, with a spatially averaged value of about 0.25 cm/yr (Delfino et al., 1993). The maximum rate of methylmercury production in peat soil, which is measured by dosing soil cores with a radioisotope of mercury, generally occurs in the 2-4 cm soil horizon (See Appendix 7-4). This is equivalent to an average age of 8 to 16 years. This zone is readily accessible to benthic infauna. Some of the inorganic mercury and methylmercury buried by depositing undecomposed plant biomass is recycled back to the surface by various physical, chemical, and biological processes or converted to elemental mercury and lost to evasion by root-mediated gas exchange (Lindberg and Meyers, 1999). The length of macrophyte roots is generally in the 10 to 30 cm range, so the mining of labile inorganic mercury and methylmercury or elemental mercury from peat soil by rooted macrophytes can occur up to depths equivalent to an average age of 120 years. However, calculations carried out by the District in last year's Chapter 7 indicate that the turnover rates of total mercury per unit area by rooted macrophytes are low compared to the annual deposition rate per unit area of total mercury by wet and dry deposition processes, even in highly eutrophic areas (pages 7-54 to 7-55 in ECR 2000). The E-MCM-II quantifies the contributions of each of these soil recycling processes to methylmercury production, transport, and bioaccumulation and predicts that the response time of the Everglades to a reduction in the input mercury load is on the order of a decade, not a century.

Comment: Has any direct relationship between P and Hg been established? If so, what is it and has it been incorporated into the E-MCM?

Response: The relationship between methylmercury in fish and water quality is a complex, multivariate one; no one variable controls mercury biotransformation and bioaccumulation across the range of conditions observed in the Everglades. Neither gross methylation of inorganic mercury nor gross demethylation of methylmercury is directly stimulated by the addition of phosphorus under the conditions studied to date (Gilmour et al., 1998a; Marvin-DiPasquale and Oremland, 1998). The correlation between phosphorus and methylmercury in fish arises indirectly from eutrophication, which induces a number of physical and chemical phenomena that influence mercury chemistry. In the Everglades, any relationship between Hg and P is complicated by other water quality variables, such as sulfur. S enrichment, also via runoff, is hypothesized to influence mercury bioavailability and biotransformation, both negatively and positively at differing concentration regimes. For example, excess sulfide produced by sulfate reducing bacteria under eutrophic conditions inhibits methylation. The E-MCM includes some features of these processes, and others, such as more detailed S chemistry, are refinements in progress. In addition, eutrophication also changes the food web and alters bioaccumulation rates. In water bodies where organic particles originate primarily with phytoplankton production and rooted macrophytes are limited to the littoral zone along the perimeter, the sorption, settling, and burial of inorganic mercury and methylmercury are dominated by primary production. Where methylmercury production is independent of the water column and sediment phosphorus concentrations, then an increase in the phosphorus load to a lake is generally associated with a decrease in methylmercury in all compartments of the ecosystem. However, phosphorus can

influence methylmercury production indirectly via its influences on the carbon, dissolved oxygen, and sulfur cycles. For example, Rudd and Turner (1983) found that methylmercury production could be stimulated to the extent that it more than offset the increase in biodilution in a particular phosphorus concentration range. Whether manipulating water column phosphorus concentrations is a viable option for managing methylmercury production and bioaccumulation was addressed on pages 7-37 to 7-39 in ECR (2000). In the northern Everglades at sites nearest District structures like those in the ENR Project and F1, the high sulfide concentrations in soil pore water are hypothesized to limit methylmercury production by limiting inorganic mercury bioavailability to sulfate-reducing bacteria (Gilmour et al., 1998b; Benoit et al., 1999). Conversely, in the southern Everglades, it has been speculated with moderate confidence that phosphorus levels fall so low that there is not enough productivity to produce the low dissolved oxygen values and carbon substrate required for active sulfate reduction and mercury methylation (USEPA, 1998). This is an area for further study.

These questions will be the subject of presentations made to the peer reviewers at the workshop.

Comment: Good luck with implementation of the biosensor bacteria – terrific technology and a terrific solution to a difficult problem.

Comment: Can SAMPEX be repeated using single rainfall events?

Response: While we agree that it is desirable to do so, in practice airborne sampling campaigns are too expensive to maintain for event sampling. As was detailed in the section on new research, DEP and EPA have set up a ground-based atmospheric monitoring site near the Everglades to collect daily samples for rainfall, fine and coarse particulates (to be analyzed for mercury & other trace elements, carbon, and particle morphology and composition), and speciation of atmospheric gaseous and particle-phase mercury. This site is scheduled to operate for 3 years and provide high-resolution data for use in receptor modeling (CMB-8 and the newer statistical models) to determine local source-receptor relationships and apportionment using multivariate trace element signatures.

Comment: Why are there Hg hotspots in parts of the Everglades not directly subject to agricultural runoff? Are precipitation patterns that predictable? Is Hg naturally concentrated by ground-based environmental factors?

Response: The Hg hotspots refer to fish bioaccumulation. Transport and deposition modeling suggests that Water Conservation Area 3 is the area most affected by Hg emissions from emissions in southern Florida. Elevated fish Hg in this region also may reflect increased net methylation rate (more production – less decomposition) or food web dynamics that facilitate bioaccumulation.

Comment: Regarding the experiment to determine the effects of mercury on wading birds, will ‘control’ birds in Tampa also receive supplemental food?

Response: Yes?

Comment: Hg research reinforces the notion that the STAs should not be permitted to dry out. Is MeHg release also facilitated by dryout of marl-based parts of the Everglades?

Response: One might speculate with moderate confidence that the more mineral marl-dominant soils may actually release a greater fraction of the bound Hg(II) than organic peat soils,

thus increasing their susceptibility to post-dryout release of Hg(II) for rapid methylation and bioaccumulation following reflooding.

Comment: Is the relationship between fish tissue MeHg and pore water MeHg strong enough for the District to encourage development of a less difficult pore water measurement technique?

Response: For research purposes, the District also uses a pore water equilibrator, which provides a better measure of the average concentrations of the truly dissolved fractions of ions that pass through the collector membrane than conventional syringe, squeeze, or porous casing collection methods. However, for more practical applications, determination of compliance with standards by using an inherently expensive and complex sampling technique like pore water monitoring is not an attractive idea. One of the tasks remaining of the SFMSP is establishing an appropriate water quality criterion and a reasonable method of ascertaining compliance.

Comment: The discussion of inflow/outflow Hg concentrations at STA-6 (page 7-26) should be expanded to provide more detail and possibly a figure illustrating the purported trend. STA-6 should afford us an opportunity to further understand Hg sequestration and transformation processes, and help us to verify/validate models.

Response: Figure added. There are no further studies planned for STA 6 at this time. Studies of the effect of extended periods of dryout and reflooding on methylmercury production and bioaccumulation will be carried out by USGS-Madison in mesocosms emplaced at WCA-2A-F1, WCA-2A-U3, WCA-3A-15, etc. and dosed with various stable isotopes of mercury and 34-S.

Comment: Data do suggest that fish tissue Hg levels are decreasing, but bird feather data appear to be too variable to draw any conclusion.

Response: Caution must be exercised when interpreting the feather data, especially in light of the observed increase in 1997 (Figure 7-2; however, note that feathers were not collected in 1996). Use of avian eggs or feathers to biomonitor differences across space or time are only useful if the observed change reflect changes in mercury contamination of the ecosystem. This relies on the assumption that the diets of the birds are constants across the same space- and time-scales. A change in diet will likely alter mercury exposure and burdens, if mercury levels vary among prey species. The great egret's diet consists of invertebrates, such as crayfish, various fish species and amphibians and differs from year to year (Frederick and Collopy 1988, Frederick et al. 1997). For example, Frederick et al. (1997) report that the proportion of warmouth (*Lepomis gulosus*) in regurgitum from chicks varied from 0.0% to 79% (i.e., % of total biomass) during the period from 1993 to 1996. Warmouth in the Everglades have been found to have high THg concentrations relative to other fish species of similar length (SFWMD, unpublished, T. Lange, FFWCC, pers. communication). Therefore, its proportion in the egret's diet may have extra significance in their exposure to mercury. Great egrets have been also called the true habitat generalists (Ogden 1999). Changes in foraging habitat, possibly due to changes in hydrology, could easily lead to a diet shift (e.g., variation in fish species among habitats, larger size fish concentrated in low water years). Such a diet shift exaggerating small differences in THg levels in the local environment, in space and time, could account for the differences in feather-THg levels between colonies and the interannual variation observed over the last five years in THg levels in egret nestling feathers. Our ability to interpret the data is improved if we focus on temporal trends at each of the colonies separately. When this is done trends become more evident.

Nevertheless, trend analysis of egg- or feather-THg concentrations should be evaluated cautiously and, where possible, should include an assessment of THg concentrations in their prey. As you note, results from fish collections by the FGFWFC, now known as the Florida Fish and

Wildlife Conservation Commission, also show a decrease in THg concentrations in fish collected at several Everglades sites over the last few years (Lange et al. 1999). Therefore, collectively these results suggest there has been a decline in levels of MeHg and possibly THg in these areas.

Comment: o In Appendix 7-3b, has the assumption that wading birds feed repeatedly in the same area been verified?

Response This is an assumption made to provide a “worst-case” analysis. In fact, the birds feed where the energy return is most favorable and that can be many locations. In practice, wading birds range over the whole of south Florida and in some cases over the whole of the Southeastern region.

Comment: The difference in opinion between the District and its partners and Exponent has persisted for at least one year. I would benefit from a concise presentation that compared the assumptions and model components from each analysis. Also, I am interested in the participants in the South Florida Mercury Science Program and the conclusions of the meeting referenced by Exponent.

Response: The participants in the South Florida Mercury Science Program include the Florida Department of Environmental Protection, the Florida Fish and Wildlife Conservation Commission, the South Florida Water Management District, USEPA Region 4, USGS, US Army Corps of Engineers-Jacksonville, NOAA, Florida Power and Light, the Florida Coordinating Group (of power companies), and the Electric Power Research Institute. Research institutions involved include the National Academy of Sciences, the Universities of Florida, Michigan, and Wisconsin, Florida International and Florida State Universities, and Oak Ridge National Laboratory. Both the District and Exponent presented their probabilistic ecological risk assessment papers at the SFMSP’s Annual Mercury Workshop at Tarpon Springs in May 2000. The peer reviewers did not explicitly address the differences in the two approaches, but Peter Frederick of the University of Florida did support the District’s interpretation and application of the relationship between fish size and foraging preference (i.e., larger birds do not forage preferentially on larger prey), and the District’s exposure and toxic effects estimates are supported by other lines of evidence.

The differences in the assumptions and data sources for the deterministic ecological risk assessments performed by the District (EIR, 1999) and Exponent (1998) are summarized on page A7-48 of Appendix 7-3 of the EIR (1999). The differences in the risk estimates for unimpacted WCA-2A-U3 and the positive control site, WCA-3A-15, have narrowed by almost an order of magnitude as Exponent has substituted Everglades-specific exposure assumptions and concentration data for default exposure assumptions and modeled concentration data. The technical differences in the way the District and Exponent treat the same Everglades-specific concentration data for their probabilistic ecological risk assessments are now more subtle and related to defining and applying appropriate probability distributions to represent each of the variables in the exposure calculation. These differences have a greater effect on the 95<sup>th</sup> percentile upper bound exposure estimates than on the corresponding 50<sup>th</sup> percentile (mean) values. The District has validated its mean estimate of methylmercury exposures to the WCA-3A colonies by comparing measured with predicted residue levels in feathers and eggs and concluded that the District’s exposure model tends to overestimate average exposures by about 30%, providing a margin of safety in the risk calculations. The differences in the mean values of the District (2000) and Exponent (2000b) risk calculations for wading birds and fish-eating mammals are now less than factor of two, while the differences in the 95<sup>th</sup> percentile upper bound values are generally within a factor of five.



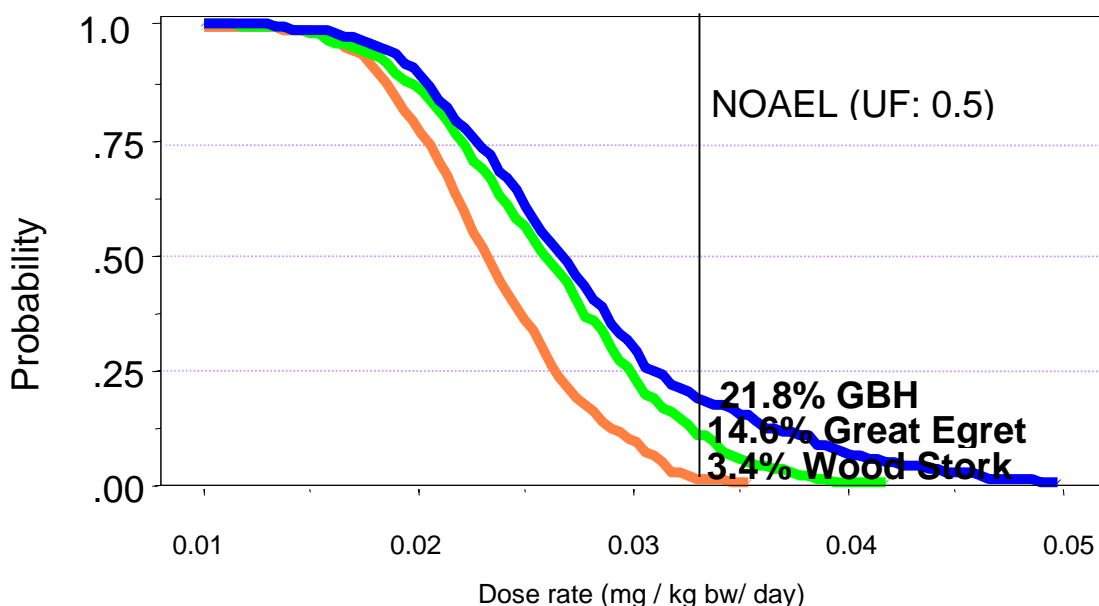
With the recent revision to Exponent's risk assessment and the reduction in their hazard quotient, which has decreased from about 10 to about 1.5 for the median exposed great blue heron (i.e., 50<sup>th</sup> percentile; Exponent, 1998; MacKay et al., 2000), the principle differences remaining between Exponent's assessment and the District's assessment are interpretation and communication of risk. Exponent has concluded from the same literature toxicology studies used by the District that wading birds exposed above the estimated no observable adverse effect level (NOAEL) will experience complete reproductive failure, while the District has concluded that only a small percentage of the birds so exposed have the potential to experience some adverse effects. To some degree this is understandable, because there are no ecological risk levels that are recognized as being either clearly acceptable (de minimis) or clearly unacceptable (de manifestis) (Suter, 1993). The USEPA guidance document (1999, pg. 5-35) states:

"In contrast to the case for human health risk assessments (where default risk-based decision guidelines are well established), there are no established default decision guidelines for identifying when risks to ecological receptors are of concern. However, if risks to nearly all members of the population (including the RME individual) are below the appropriate NOAEL-based toxicity reference value, then it is likely that risks are within an acceptable range both for individuals and the population. Conversely, if risks to the average members of a population exceed an appropriate LOAEL-based TRV, then it is likely that risks are not acceptable, either to individuals or the population."

The District's probabilistic risk assessment predicted 78.2% of the modeled great blue heron population at the Post-ECP reference site would receive a daily dose rate below the estimated no-observed-adverse-effect level (NOAEL, Fig. 1). Hence, this segment of the population is believed to have no appreciable risk of deleterious effects from mercury exposures.

However, **Figure 1** indicates also that 21.8% of the population is expected to receive a daily dose rate exceeding the NOAEL and, thus, are at some elevated risk. That is not to say that 21.8% of the population will suffer reproductive failure.

To assess the biological significance of the risk to this great blue heron population, one must first understand the basis of the LOAEL and the derived NOAEL-based toxicity reference value (TRV) that was used to evaluate the exposure distribution. This includes the nature of the LOAEL endpoint, its relevance to the assessment endpoint, and the shape (steepness) of the dose-response curve (USEPA, 1999).



**Figure 1.** Reverse cumulative density function of dose rate for mercury exposure to wading birds feeding exclusively at the Post-ECP reference site (Fig. 7; Rumbold, 2000). Note, the lowest-observed-adverse-effect level (LOAEL), 0.064 mg/kg-bw/day, not shown on x-axis.

As discussed in Appendix 7.3b (Rumbold, 2000a), the USEPA derived the LOAEL from a series of studies carried out by Gary Heinz (USEPA, 1993; 1995). Heinz dosed three generations of mallard duck hens ( $n = 74$ ) at 0.5 mg MeHg/kg food eaten. During the first generation study, Heinz (1979) determined that 0.5 mg MeHg/kg food had no significant effect on reproductive effects such as hatching success or duckling survival (i.e., no-observed-adverse-effect-level or NOAEL; dose rate of @ 0.064 mg/kg-bw/day; see Rumbold, 2000a regarding calculation). However, second generation dosed birds (and when data were pooled from all three generations) showed significant differences from controls in terms of:

- Percent of eggs laid outside nestbox
- Percent of sound eggs
- Number of 1-week-old nestlings produced
- Percent of ducklings approaching calls
- Distance traveled in avoidance

Because the study included only a single dosing level, Heinz (1979) did not develop a dose-response curve for any of the measured effects. While the differences between the dosed and control birds were statistically significant, the size of the average effect was often small. For example, in terms of mean measurements from the three generations combined:

- Percent of eggs laid outside the nestbox increased from 4.3% in the controls to 9.7% in the dosed group.
- Percent of sound eggs decreased from 0.69 to 0.59 eggs laid per hen per day in the dosed group.

- Number of 1-week-old nestlings produced decreased from 46 in the controls to 37.5 in the dosed group.
- Percent of ducklings approaching calls decreased from 97% in the controls to 94% in the dosed group.
- Distance traveled in avoidance increased from 32 cm in the controls to 38 cm in the dosed group.

While it is clear that some of the mallards suffered decreased reproductive success, the mallards did not suffer complete reproductive failure when dosed at the LOAEL. Accordingly, it would be incorrect to infer that 21.8% of the great blue heron population at the Post-ECP reference site will suffer reproductive failure because their exposure exceeded the estimated NOAEL, which is  $\frac{1}{2}$  the dose rate of the mallard LOAEL (cf. MacKay et al., 2000). Instead, the only conclusion that can be drawn from the available data is that these birds are at elevated risk of some adverse effect.

The District's assessment attempted to provide perspective for assessing this risk using a positive control site, i.e., the mercury "Hot Spot" at WCA-3A 15. Despite a much higher risk level (i.e., > 99% exceeded NOAEL and 83.8% exceeded LOAEL; see Fig. 8 – Rumbold, 2000a), no evidence of impaired reproduction (i.e., fledging success) has been reported for wading birds nesting near the positive control site (for review see Rumbold, 2000a). In fact, quite the opposite has been reported. The 3-year running average for great egret nests in the Everglades has increased since the mid-1980s and peaked in 1999 (1999 nesting approached record nesting observed in 1940s; Ogden, 1999). Further, clutch size and more importantly nest success for great egret colonies in WCA-3A have recently been the highest recorded for the Everglades and in the upper range for the species, overall (Frederick et al., 1997). As previously mentioned, the District also validated its exposure model by predicting concentrations of mercury in great egret eggs and feathers collected from colonies near the positive control site. The results of the validation exercise showed that levels observed in eggs and feathers were less than model-predicted concentrations and suggest that the 1-D MCA overestimated mercury exposure to the birds. More importantly, observed tissue concentrations near the "Hot Spot" were also below levels thought to be of toxicological concern (Rumbold, 2000c).

As evident from this discussion, in accordance with USEPA guidelines for ecological risk assessment (1998), the District did not rely solely on a mathematical exposure model in its assessment of risk at WCA-2A U3, but instead made its determination based on multiple lines of evidence (cf. MacKay et al., 2000). Appendix 7-3b in ECR (2000) sets forth all of the data, assumptions, approximations, and extrapolations that were used by the District in developing and applying its exposure and toxicity models, the exposure validation analysis, and the toxicity validation analysis for its probabilistic ecological risk assessment for wading birds. Since the District was unable to extract this same information from the most recent submittal by Exponent (MacKay et al., 2000), we are unable to make the requested comparison.

Comment: Exponent should be prepared to discuss whether the relationship between P and Hg is coincidental (correlation) or actual (regression), and the costs and benefits of continued pollution of the Everglades.

## Chapter 7: The Everglades Mercury Problem

### Responses to Public and Peer Review Panel Comments

By Tom Atkeson & Paul Parks of DEP and Larry Fink &  
Darren Rumbold of SFWMD

William Green, Esq., Sugar Cane Growers Cooperative of  
Florida, Comments and Author's Responses

Comment: An independent advantage of allowing for gradient ecology is that such areas currently have the lowest methyl mercury contamination levels, attributed in part to higher phosphorus levels seen in those areas.

Response (DEP, Atkeson & Parks): The Everglades Forever Act (EFA) requires that the impacted areas proposed by Mr. Green for “gradient ecology” be restored to Everglades habitat typical of the area and hydroperiod. When this is done, phosphorus levels will fall and prey fish mercury levels are expected to rise to values typical for the area and hydroperiod. Gradient ecology in the impacted areas would mean higher phosphorus levels and potentially somewhat lower prey fish mercury levels compared to the EFA requirements. It is to the lower prey fish mercury levels of gradient ecology that Mr. Green attributes an advantage, i.e. lower mercury exposure to predators such as wading birds that consume the prey fish in these areas.

DEP agrees with Mr. Green that wading birds feeding exclusively in the restored areas are likely to be exposed to less dietary methylmercury under the gradient ecology regime than under the EFA requirement. However, because the exposure of wading birds in areas restored to the EFA requirement would not rise to the level of harm, DEP believes that the advantage conferred by the lower exposure of gradient ecology is not significant. The DEP reasoning on this is as follows.

Some increase in prey fish mercury in the previously impacted areas is not necessarily harmful. The significance of the increased exposure must be determined by an accepted technique for risk analysis. Risk is determined by comparing a daily reference dose of mercury called the No Observed Effect Level (NOEL) with the daily dietary intake of mercury. The actual daily dietary intake of mercury cannot be determined for wading birds feeding in the post-restoration impacted areas because restoration has not yet taken place. The best approximation is wading bird mercury exposure in an existing area of the Everglades that closely resembles the habitat expected for the impacted area after restoration, a technique often used and referred to as the ‘reference site’ approach. Resemblance is important because the Everglades is not homogeneous with respect to mercury.

In its ecological risk assessment, SFWMD has taken this approach (Everglades Consolidated Report 2000, Appendix 7-3b). SFWMD determined the mercury exposure of wading birds feeding at a reference area that is expected to be like the impacted areas after restoration to EFA requirements. In the SFWMD study, the risk to wading birds feeding exclusively at the reference site was not unacceptable when actual exposure was compared to the NOEL determined on a study of tame mallard ducks (a NOEL for wading bird species is not available). By inference, the same exposure to mercury and the same lack of significant harm would occur for wading birds feeding exclusively at the impacted areas after restoration. SFWMD concluded:

“Although certainly not trivial, the baseline methylmercury risks to wading birds feeding exclusively at WCA-2A-U3 (i.e., reference site for Post-ECP conditions) are considered acceptable. Therefore, restoring nutrient impacted areas in the northern WCA’s to the conditions which now exist at WCA-2A-U3 are unlikely to be a threat to wading birds feeding in those areas, i.e., the methylmercury risks will not be unacceptable.”

DEP concurs that the reference site chosen by SFWMD, the prey fish mercury levels used to compute exposure and the risk assessment techniques used by SFWMD represent the best available information for this comparison. The Peer Review Panel for the ECR2001 (this volume) had this to say about the SFWMD analysis:

“The SFWMD is to be commended for their use of probabilistic risk assessment for the wading birds. This is the current cutting edge risk methodology, and the assumptions and parameters used in these assessments are sound and reasonable. While the NOEL may be high because of its development for Mallards, it is the conservative approach until another NOEL can be developed that is more appropriate. A sensitivity analysis may indicate what factors are most subject to having an effect on the affects endpoints.”

As noted by the Peer Review Panel for this report, the NOEL for wading birds is likely to be higher for wading birds than for Mallard ducks. If true, this means that the actual risk is lower than that calculated by SFWMD.

As discussed in Chapter 7 of ECR2001, trend monitoring data for Everglades fish and birds suggest declining mercury contamination. While this evidence is somewhat preliminary, it is consistent with the time lag predicted by modeling for a decline in atmospheric deposition resulting from decreasing amounts of mercury emitted by air sources within south Florida. Further declines in wildlife mercury exposure from these control measures are possible. Additional controls may be possible and may produce more reduction in exposure. DEP believes that it is premature to rule out the possibility that emissions controls can further reduce exposures in the entire Everglades including the impacted areas for which Mr. Green has proposed gradient ecology.

Comment: We also suggest that caution be applied to the research hypothesis that EAA sulfate may enhance mercury methylation at certain concentrations in certain areas of the Everglades. While hypotheses such as that formulated by Dr. Gilmour are a valuable component of scientific research, they should not be assumed true until proven. This is especially the case for the sulfate hypothesis which is both contrary to what has been observed in other wetlands and is based on limited data over a wide geographical region. In contrast, an inverse relationship of

phosphorus and mercury in fish is apparent in water bodies throughout the country and other parts of the world, such as Scandinavia.

Response (DEP, Atkeson & Parks): Atmospheric deposition is the source of most of the mercury that impacts Everglades biota. Once deposited on the marsh, ubiquitous sulfate reducing bacteria convert inorganic mercury to methylmercury, a form that can be magnified to toxic levels in Everglades predators such as wading birds. From intensive field investigations and computer modeling studies, DEP has found that fish mercury levels are nearly directly proportional to the rate of deposition of mercury from the atmosphere. Indeed, field data on largemouth bass and wading birds indicate a declining trend in mercury concentration that is likely attributable to declining mercury emissions from sources within south Florida. Because mercury emissions to the atmosphere are the first step in creating the mercury problem, DEP's primary strategy for control is emissions reduction.

If it becomes apparent that emissions controls are not sufficient to reduce Everglades mercury to acceptable levels, DEP will necessarily turn to the second step, devising management steps to limit the production of methylmercury. At present, there is no evidence that this is possible, but there is suggestive evidence that it may be. In the event that emissions controls are insufficient, a candidate for further investigation is control of sulfate in EAA runoff. It is not known at present how strongly the present-day range in concentration of Everglades sulfate may contribute to mercury methylation nor whether sulfate control is a feasible technique for alleviating the Everglades mercury problem.

Long-term trend data for mercury in Everglades biota are strongly suggestive of a declining trend in largemouth bass and wading birds in the late 1990's. There is cause for cautious optimism that this downward trend will continue. It may be that emissions control measures already taken will reduce Everglades mercury to acceptable levels. DEP, EPA, USGS and others continue to work to determine what level of emissions control is sufficient. This necessarily involves work on the processes that govern methylmercury production and bioaccumulation. From the process work, DEP expects to understand the effects of sulfate concentration more completely. At present, DEP has no clear evidence that control of sulfate in EAA runoff has the potential to alleviate methylmercury bioaccumulation in Everglades biota. If evidence comes to light that control of EAA sulfate is both necessary and efficacious, DEP would begin to work toward that end in cooperation with EAA growers. In the meantime, DEP has no plan or intention to institute such controls.

## Chapter 7: The Everglades Mercury Problem

### Responses to Public and Peer Review Panel Comments

By Tom Atkeson & Paul Parks of DEP and Larry Fink &  
Darren Rumbold of SFWMD

The first section consists of a list of Panel organizational and presentation recommendations for the 2002 Everglades Consolidated Report and future reports. These are accepted and will be undertaken in the future. No more specific response is necessary.

The second sections consists of panel questions, comments and suggestions for future research. There is a specific response for each subject area.

The third section consists of panel suggestions for improvement in the ECR 2001 Chapter 7 or its Appendices. These are listed with a reference to the changes that were made. If changes were not made, the reason is given.

In all three sections, where comments concern the same subject area, they are grouped together. Page numbers preceding comments refer to the Peer Review Panel Report.

#### A List of Organizational and Presentation Recommendations for the Future.

Comment (Page 2): Regarding the material in the chapters, the panel notes that a number of issues are common among many chapters. For example, there are two to four major issues with mercury in the Everglades that are also seen in other chapters. It would be useful to organize the introductory material to address these issues, with the appendices grouped to reflect these themes. The panel feels a better job could be done to coordinate inter-chapter and appendix material. Rather than treat each chapter as an individual document, the coordinator of the whole Report should be given more authority to integrate the document.

Comment (Page 2): Finally, regarding the review process, for complicated topics such as mercury, individual secondary reviewers should be assigned specific issues (such as atmosphere cycling, internal cycling, fate and biological effects, and modeling).

Comment (Page 2): As the science behind future criterion development unfolds, it would be extremely helpful to future monitoring, data analysis and management decision making to have scientifically defined violation definitions of standards built into the criteria. A 'template' for developing a peer reviewed definition of P criterion compliance may be that used in the dissolved oxygen criterion evaluation. We view dissolved oxygen violation definition in the criterion evaluation a critical component for connecting monitoring results to management decision-making.

Comment (Page 19): The report should attempt to define "restoration" or "recovery" in a way that relates specific management goals or legislative mandates of P or Hg thresholds and levels to the more general goals of the CERP and the long-term health of the Everglades system. This should be done even if a range of P level is eventually considered as a management goal with a corresponding range in the impact of these management options to what is considered to be "restored." Perhaps some attempt should also be made to distinguish between the potential impact of P and Hg in the overall management scenario.

Comment (Page 19): It seems logical that as investments are initiated, a summary, perhaps in a tabular format, be included at the outset of this chapter. This table should also include some statement as the projects relate to issues such as management of TP, site specific P, Hg, etc.

Comment (Page 29): Chapter 4 in addressing standard compliance, presents a major line of evidence demonstrating management's accountability for the water quality 'goals' established for the Everglades. The tables of standard violations attempt to summarize large volumes of data into a digestible format. The discussions of standard violations, organized by water quality constituent, provide additional insight into the nature of the issues facing water quality managers in the Everglades. The appendix presenting the dissolved oxygen standard analysis is well done and documented as is the appendix addressing chronic toxicity based guidelines for pesticides and priority pollutants.

## Panel Questions, Comments and Suggestions for Future Research

Comment (Page 9): The marsh dryout studies will contribute greatly in predicting the impact of a drought. The results suggest that dryout should be avoided at all costs. Have the short- and long-term impacts to downstream systems been considered if dryout cannot be avoided?

Response: The natural Everglades experienced frequent extended periods and areas of dryout and occasional plant top fires, so suppressing this natural phenomenon to reduce the magnitude of the spike in MeHg production would appear inappropriate. Source reduction will reduce the reservoir of available Hg(II) in soil over time, so that the occasional dryout and fire will result in a lower peak. The modifications of E-MCM-2 should make it possible to evaluate the sensitivity and relaxation time of the mercury cycle in response to changes in various environmental parameters, such as water stage-duration, total phosphorus concentration, and sulfate concentration.

Comment (Page 16): Chemical Treatment-Solids Separation- The pH value and alkalinity can have a pronounced effect on algae growth. Bioassay result will give some indication of what impact might occur, but has any thought been given to what impact the discharge of the treated water to a natural environment might have on periphyton, plants, etc.? With increases in sulfur concentrations is there a potential for blooms of sulfur bacteria and an impact on the production of methyl mercury from the deposits of particulate mercury?

Response: The concentrations of total phosphorus in these areas should be insufficient to foster the conditions that favor the thick mats of spirogyra and low D.O. that favor the presence of sulfur and sulfate-reducing bacterial communities and foster MeHg production. Nevertheless, sulfide is toxic to both aquatic plants and animals in sufficient concentrations, and the potential for such toxic effects need to be taken into account prior to creating such discharges. Other questions about the effects of treatment in removing metals and changing ionic strength are under consideration for CTSS.



Comment (Page 34): One overall goal is to arrive at a suitable criterion that will protect the health of the biota residing within the system, including humans.

Response: Developing a more appropriate mercury criterion is a requirement of the Everglades Forever Act and is prerequisite to defining the controls necessary to alleviate the Everglades mercury problem.

Comment (Page 35): The report indicates that 95 % to 98 % of the input of Hg is from atmospheric deposition, and that only 5% comes from local sources by inflow from upstream areas. The important question, however, is the relative contribution of local sources of atmospheric Hg compared to global sources. If atmospheric Hg is mostly global, it limits the potential reductions in the Hg that will result from further reductions in local sources. It would be easier to evaluate the relative contribution of local vs global sources of atmospheric Hg if there were one clear section devoted entirely to showing the data and models, and assumptions, made to derive this estimate. While there are a number of appendices devoted partially to this problem, the data necessary to evaluate this aspect are not presented in the current report. This information includes: the assumptions inherent in these models, the sampling regime implemented during all months, and the lack of data on atmospheric deposition data for several years. Some of this information was provided in presentation at the meetings, but was not presented in enough detail or in written form, making it difficult at present. These data, however, are extremely important, and should be presented in one clear section in future reports.

Comment (Page 35): The University of Michigan models as presented are both useful and laudable, but the data presented in appendices seem to concentrate on limited field work. Presentations at the Workshop made it clear that extensive field data are available, and these should be incorporated in future reports. These data would be easier to evaluate if there were one detailed section on mercury inputs to the Everglades.

Comment (Page 35): In any event, it is not known what fraction of atmospheric Hg is from local and regional (within Florida) sources, which presumably could be controlled, and what fraction comes from truly global sources, which will be harder to control. The decline in Hg to the Everglades fish and bird feathers over the last several years may partially result from reductions in local and regional air emissions. Monitoring should be instituted to determine the percentage of atmospheric Hg that is due to local and regional atmospheric inputs, particularly from the east coast of Florida.

Comment (Page 35): Finally, there is considerable concern in the Northeast that energy deregulation and the increased use of coal will result in increased levels of atmospheric Hg. Deregulation of energy may increase the demand for cheap, coal-based electricity in plants whose environmental controls are marginal, thereby increasing the overall emissions of Hg to the atmosphere. Whether this would reach Florida, lying in a prevailing easterly zone is uncertain. While information presented at the Workshop suggests that 20 % or less of the atmospheric Hg wet deposition comes from non-regional sources, this needs to be considered. It is critical to obtain more data on rates and the importance of dry deposition to the Everglades.

Response: DEP is aware of the necessity of distinguishing controllable from non-abatable atmospheric sources of reactive gaseous mercury and it was toward that end that the Speciated Atmospheric Mercury Profiling Experiment flights were directed. Reports of this experiment are expected in late 2001 or early 2002. Understanding the relationships between air source emissions, atmospheric transport and chemistry, and depositional processes, and representation of same in an advanced meteorological model are areas of active continuing work. Present plans are to complete these elements of work within about 3 years.

Although it does not figure prominently in this report, it should be kept in mind that the atmospheric elements of the SFMSP have been closely coordinated with USEPA ORD-National Exposure Research Laboratory. EPA has completed a National Mercury Research Strategy that includes substantial work on the atmospheric mercury cycle, which should complement and greatly enhance the work done within Florida, particularly with respect to the larger scale aspects of transport. An early example of this is the Mercury Super Site described in the chapter that will build a long-term, high-resolution record of mercury and other atmospheric measurements for use in modeling. Should EPA proposals mature, we hope to follow up with a 3<sup>rd</sup> generation source-receptor study in southern Florida that would address the concerns expressed about the short duration of SoFAMMS..

The pilot TMDL study from which the source-receptor are derived is under revisions following peer review. In that revision, the data, models and assumption will be presented in a more understandable fashion. The results will be presented in the 2002 edition of this report.

Work is in progress on dry deposition rates. See Appendix 7-5.

Comment (Page 35): It would be useful to know whether agricultural sources of Hg have been examined. For example, in some places Hg was used in agriculture as a seed dressing.

Response: See Responses to Questions Asked Prior to the October 6, Peer Review Panel Workshop by Dr. Joanna Burger.

Comment (Page 35): Several relationships about Hg cycling within the Everglades are apparent from reading the report and appendices, but some of these require further explanation and development. While the data are in the appendices, they could be more easily interpreted if presented in one place. These include the relative relationships and the role of links between: a) construction work and MeHg levels in fish and birds, b) drying and reflooding caused by La Nina in 1999 and the increase in MeHg in surface water, c) initial flooding of STAs and flushing of MeHg and total Hg from the peat soils, d) total Hg and MeHg during pre and post-flooding events, e) fluxes of total Hg and MeHg within the STAs, f) bioaccumulation factors in different regions of the Everglades, and g) sulfur and methylation. The importance of maintaining sufficient water levels in the STAs so that there is not drying and reflooding, which increases methylation, is a critical point worth developing further.

Response: DEP intends to use the E-MCM to integrate the processes affecting methylmercury bioaccumulation. DEP is aware of the need for a better way to set forth the components of the conceptual module, the field results that underlie them and lacking information that must be from ongoing research. Necessary, but unfunded research must also be included in this exposition. Work is in progress toward that end and results will be reported in ECR 2002.

Sufficient field data are not available for all of the processes mentioned above, e.g., drying and reflooding, to apply these processes to the model. Until all significant processes are incorporated, model application relies on calibration, which, as discussed elsewhere, is not sufficiently constrained.

Comment (Page 35): While the overall cycling of Hg in various forms is of interest to understanding the Hg problem in the Everglades, from the viewpoint of ecological receptors, the conversion of elemental Hg to MeHg is the key step. It is also the step that is controversial because of the many factors that affect methylation rates. Further, since Hg can be sequestered in sediments, it is available for later resuspension and methylation long after water Hg levels appear low (particularly if bottom sediments are disturbed).

Response: Except for sulfide concentration, the factors determining the availability of Hg(II) to sulfate reducing bacteria are largely unknown. An additional complexity is that some of the factors that affect methylation rates, also affect food web dynamics, e.g., productivity and dissolved oxygen.

Comment (Page 36): Hg cycling within the Everglades: It appears that nearly all of the Hg deposited to the Everglades is retained and accumulates in the peat and sediments. Any disturbance of this pool, whether by mechanical or hydrological effects, can flush out total Hg and MeHg. It is this internal cycling of total Hg and MeHg which is critical and which bears further investigations. The efforts of the SFWMD are clearly directed toward understanding these relationships, and this understanding is guiding their management and restoration efforts, to their credit.

Response: The mesocosm studies are underway that attempt to discriminate the peat soil reservoir contribution to the inorganic mercury flux and methylmercury production as a function of dryout, water depth, and water and sediment chemistries. Results will be reported in ECR 2002.

Comment (Page 36): The relationship between P and Hg methylation, discussed more strongly in previous reports, is complex. The suggestion is that with increases in P and eutrophication, plant production increases, standing crop increases, and decomposition increases, which leads to higher production at all trophic levels. This leads to biodilution of the toxicants. There are many problems with this, including a lack of a tight relationship between P and methylation, even with biodilution the Hg remains in the system and can continue to be cycled, and methylation is associated with other biogeochemical factors such as C and Fe. Gilmour and Krabbenhoft found no direct effect of phosphate and nitrate on MeHg production rates in sediment cores.

Response: Ongoing mesocosm studies will help clarify this. Where there is eutrophication, sulfate is present in great excess. The effects of eutrophication on sulfide relate P to methylation, but in ways not well understood. Bioaccumulation is dependent upon food web dynamics as well as upon the net rate of methylmercury production.

Comment (Page 37): Given that MeHg occurs, there are two other questions that are critical to understanding (and thus managing) the Hg problem in the Everglades: what factors affect bioavailability, and how does the MeHg enter the food chain? It would be useful to see some data on the levels of selenium in the Everglades, particularly in the regions of high, medium and low MeHg levels. Selenium is known to reduce the absorption of MeHg mercury in vertebrates, and to partially ameliorate the effects of MeHg. Thus, to understand the effects of MeHg it is essential to know the levels of both in biota.

Response: Current knowledge about the entry of methylmercury into the food web is summarized in Appendix 7-4. It appears that uptake by benthic organisms is the primary route in the marsh. No aspect of selenium interaction with mercury has been examined for the Everglades. Selenium is assumed to be an invariant part of the background. Selenium would be important, if it exerted different effects depending upon location or conditions. However, resources to investigate it are lacking.

Comment (Page 36): The effect of drying events and fire are increasing the concentrations of MeHg, and need to be examined more closely with respect to Hg cycling, and to management of the STAs. Drying events, followed by flooding, appear to release a massive pulse of MeHg production, and may provide a useful method to quantify how hydrologic flow pathways as well

as how the sulfate/sulfide balance controls methylation, and should be explored whenever the opportunity permits. It also suggests that the effects of drying and flooding events and fire need to be factored into the mercury cycling models for the Everglades.

Comment (Page 41): Determining the input sources from atmospheric and old accumulated Hg and MeHg in peat to the Everglades is a high priority. Understanding the variations in the contribution of local and global atmospheric Hg seasonally and annually are critical to the models for Hg inputs into the Everglades. Since there is the possibility of control of Hg from local sources, the relative contribution of local sources should be examined with respect to spatial, seasonal, and annual patterns.

Response: Because they are closely related to Everglades water management, which is fundamental to the restoration effort, the effects of drying and reflooding on methylmercury production and uptake are of great importance. Mesocosm studies now underway will help answer these questions. Those results will be reported in ECR 2002. The E-MCM-2 is a dynamic model and the modeled rate of MeHg production will respond to a shift in the pore water concentrations from organic sulfide to inorganic sulfate following a simulated dryout event. More research may be needed to calibrate and verify this aspect of the model.

Comment (Page 36): Methylated Hg leaves the sediments (and the periphyton layer) by solute efflux from the sediment porewaters, by movement of benthic invertebrates into the water (or water column), and by direct grazing on surface sediments, benthic invertebrates, and the periphyton. The finding that methylation is very rapid in periphyton is extremely interesting and potentially important because it provides a rapid and direct method of entry into the aquatic food web, leading directly to higher trophic level fish, birds and mammals.

Comment (Page 43): The relatively recent finding of the importance of the periphyton to methylation needs to be further examined and studied to understand its contribution to the food chain (relative to surface sediments). The temporal and spatial factors that affect the relative percentage that periphyton contributes to overall MeHg in the Everglades should be examined over enough seasons to understand fully its importance.

Response: Methylation of mercury is not rapid in the type of periphyton community characteristic of the oligotrophic Everglades. Only in the eutrophic areas are there periphyton communities where mercury methylation is rapid.

Comment (Page 38): It is critical that the E-MCM model is used as a research tool to help test the hypotheses being put forward to explain issues. In that sense, the changes being made to the model to link the phosphorus transport and fate model and to incorporate sulfur state variables and associated processes into the mercury model are critical. Then the refined E-MCM needs to be used in a research mode to test the hypotheses in the context of all the other forcing functions and competing processes. Among the types of observations amenable to hypothesis testing include the effect of drying/burning on mercury cycling and MeHg production in the system; the importance of diel vertical migration of phytoplankton and zooplankton on food chain bioaccumulation of MeHg; the role of sulfur transport and cycling in explaining the north-to-south gradient of MeHg (especially the "hotspot" in WCA3) and associated bioaccumulation; the linkage between Hg emissions, Hg deposition, and aquatic system response in terms of the impact and timing of reduction of local emissions on fish uptake.

Comment (Page 43): The development of a quantitative understanding of transport, fate, bioaccumulation and effects of Hg in the entire Everglades Protection Area requires a collaborative and interactive relationship between process experimentation (provides

understanding and parameterization for model development), field monitoring data (provides input and credibility for models), and modeling (provides insights and makes projections). The Mercury Science Program is strongly encouraged to continue and even enhance this relationship by using the E-MCM model to synthesize new findings and to field test working hypotheses at the whole system level.

Response: The E-MCM is intended as a tool for management decision-making. To be useful for this purpose, its process modules and their calibration and verification must be established by field data. The interaction has worked in both directions in the past. Modeling results make the need for particular field work apparent and field results make clear the need for incorporation of new processes into the model. Because of the collegial coordination of the South Florida Mercury Science Program, the close working relationship between model developers and researchers is unparalleled. If the researchers pose hypotheses, the model can be run to test them. There is sufficient flexibility in the contract to ensure that this occurs. If there is disagreement between the model and the hypothesis, follow-up research is conducted to clarify the process to fix the model or reject the hypothesis.

Comment (Page 35): There is one more important point regarding use of the model as a management tool to make forecast on future conditions in the system in response to alternate management actions (e.g., Hg emission reductions, phosphorus control, alteration of hydroperiods, reduction of sulfur transport from the EAA). Currently the model has been calibrated to a relatively small area (around WCA3-15) and over a relatively short period of time during which calibration data have been collected. The problem is that this model - and the modelers readily admit it - is underconstrained by this calibration data set; therefore, there is not a unique set of coefficients that can be used to affect a calibration. Unfortunately, the alteration of these coefficients (particularly the ones that affect long-term burial of Hg in the system) while still meeting short-term calibration targets can have a major effect on the long-term response of the system to external perturbations or remediation efforts. For this reason, it is my recommendation that a long-term hindcast calibration needs to be attempted with this model. To accomplish this, the modeling team will have to attempt, albeit difficult, to reconstruct the atmospheric loading history and other forcing functions (e.g., hydrology) over about a twenty year period during which we have fish and bird feather or egg mercury trend data to which the model can be compared. This exercise will go a long way toward increasing our level of confidence in using the model as both a management and a research tool.

Comment (Page 42): The existing calibration of the E-MCM is insufficient for it to be used as a management tool for making assessments relative to management actions in the EPA (e.g., impact of reduced Hg emission, impact of phosphorus control measures, impact of sulfur reductions). The modeling team should strive to conduct a long-term hindcast calibration (at least 20 years) and to field test the model calibration at a variety of sites within the EPA (STAs, enriched portion of WCAs, Loxahatchee National Wildlife Refuge, and ENP).

Response: The model is also being calibrated to the ENR Project to evaluate the effect of operational variables (i.e., water depth, HLR, and TP) on THg and MeHg removal from the water column, storage, and recycling; F1 and U3 along the WCA-2A transect to evaluate the effect of TP reduction on MeHg production and bioaccumulation; and WCA3A-15 to evaluate the effect of atmospheric deposition reduction on MeHg production and bioaccumulation.

A hindcasting initiative is now under way. Work is in progress on new sediment cores that may show the decline in deposition that should have occurred with the decline in emissions, the hypothesized cause of the declining trend in largemouth bass and wading birds. Hindcasting over this period would be a critical test of the model.

Comment (Page 39): Risk assessments for wildlife are difficult for several reasons. In general, effects research usually involves controlled laboratory conditions, which limit the species of birds that can be used. Much of the effects work with Hg involved ducks, which appear to be more sensitive than wading birds and seabirds. While all researchers in this area recognize the problems with using LOELs and NOELs derived from Mallards in laboratory experiments, they remain the only currently available points. The SFWMD is encouraged to continue working with researchers to develop LOELs and NOELs with more relevant species, including wading birds.

Comment (Page 39): The finding that there is no obvious effect on reproductive success in wading birds in areas of high and low Hg levels does not answer the question about early developmental effects. While it is difficult to examine the effect of mercury on chicks from day one to fourteen, experiments during this critical period would help answer the question about effects.

Comment (Page 40): The research program of Spaulding and Frederick (among others) with wading birds is excellent, but such studies are time-consuming and involve many years before all relevant aspects can all be examined. The plan of the SFWMD to examine embryonic stages and developmental abnormalities is sound in light of research with other neurotoxicants, but these studies should be conducted in the field (see above) as well as at Patuxent. Further, the plan to examine effects of Hg on adult reproductive success is also essential, but some thought should be given to examining all endpoints in the same species. That is, all endpoints should be examined in White Ibises, Snowy Egrets and Great Egrets, not just some endpoints in each. Further, one might consider examining some of these endpoints in Anhinga that live within these systems to a greater degree. It is possible to have birds with the highest levels of Hg be adversely affected while populations levels of the species overall are stable or increasing. Since many factors affect reproductive success (such as inclement weather stresses, food scarcity, predators, human disturbance), population trends in themselves are not the only measure to use in examining the effects of Hg. Further, in the field, the effects of Hg might be either enhanced or reduced by interactions with other contaminants (which needs to be examined, but little is known about this from anywhere)

Response: There are no funds for wading bird studies other than the ongoing work described in this Report. Due to a change in priorities, the planned SFWMD work is not funded. The fundamental importance of wading bird LOELs is recognized. Mike Myers' work on the loon and Gary Heinz's work on the CALFED project will be monitored.

Comment (Page 40): The planned experiments with dosed wild Great Egrets, whereby fledglings will be followed by telemetry, should yield very important and key data. It is essential that these studies have appropriate controls that experience the same initial sham dosing regime, radiotelemetry, and post-fledging dosing. While the research work undertaken to date with wading birds is excellent, and of high quality, there are other endpoints to consider, including:

- Wood Storks - of interest because they are endangered, are a key species in the Everglades, respond to management practices, and they feed on large fish.
- Raptors have proven to be sensitive to contaminants, and Osprey feed on relatively large fish. They might be expected to accumulate high levels, have long lives, and are high on the food chain.
- Mammals, such as bats, may provide some useful information as they have been shown to bioaccumulate Hg in other studies.
- The possibility that wading birds demethylate Hg should be examined in wading birds, as a possible mechanism that explains the lack of population effects in wading birds. This would involve examining the MeHg/inorganic Hg levels in liver of wading birds.

Comment (Page 43): While the research on wading birds is laudable, and clearly very important to our understanding of Hg fate and effects, there are some additional data needed to evaluate the effects on birds more clearly, including 1) radio-tracking data on the whereabouts of wading birds throughout the year, and the relative importance of the "hot spots" to yearly foraging patterns, 2) comparable data on Hg fate and effects during the embryonic and early life stages of egrets, and anhinga (a species that eats relatively large fish), and 3) the fate and effects of Hg in eggs, young and adults following massive events (such as fire, very low water).

Response: While clearly worthwhile, these studies must compete for limited funding with projects that have a very high priority of being directly involved in management decisions.

Comment (Page 41): The SFWMD is to be commended for their use of probabilistic risk assessment for the wading birds. This is the current cutting edge risk methodology, and the assumptions and parameters used in these assessments are sound and reasonable. While the NOEL may be high because of its development for Mallards, it is the conservative approach until another NOEL can be developed that is more appropriate. A sensitivity analysis may indicate what factors are most subject to having an effect on the affects endpoints.

Comment (Page 41): It is gratifying to see the growth and improvements in the Probabilistic Risk Assessment Model since it was first introduced last year. but its dependence on the mallard derived LOAEL and NOAEL is still a weak link. To the extent possible, and until better numbers become available, this input should be modified in the light of M. Meyer's loon findings, and the Frederick and Spalding study. The use of a threshold LOAEL & NOAEL should be replaced by a dose response value in the risk assessment calculation. Nichols (1999) article describing the development of the USEPA Wildlife Criteria may be useful in this context.

Response: Due to a change in funding priorities, SFWMD is no longer involved in risk assessment or any other aspect of Everglades mercury research. DEP does not have current funds to extend this work.

Comment (Page 41): It is interesting to learn that wading bird breeding success has been better this year than in most preceeding years for which data are available. It was stated during the presentations on Thursday that Peter Frederick speculated that increased nesting success could be related to decreased tissue MeHg. This is consistent with the findings of Barr and of Heinz et al, also Nocera and Tayloer (1998) on MeHg- induced impairments in reproductive/parental behavior, but one would not expect an effect to show up in the same year. When did the fish tissue MeHg concentration first begin and how long has this trend been observed?

Response: Apparently, this comment was made by someone other than Peter Frederick. Everglades wading bird nesting success is closely related to antecedent and current water levels. While lower mercury levels may have been a factor in nesting success, there is no evidence that this is so; nor, is there any evidence that mercury has been responsible for reduced nesting success in the past.

Comment (Page 41): Potential damage to bats should always be considered when assessing risk or deriving standards for waterborne contaminants, especially those that bioaccumulate. Bats are long-lived and have low reproductive rates. Although MeHg content of emergent aquatic insects is much lower then MeHg content of fish at the same site, bat insect ingestion rates are high (wet weight of insects on the order of 0.5x-1.0X the bat's body weight/night) so that the potential for contaminant exposure and accumulation via the food chain is high. For example, a bat of 10 grams body weight, and 5 to 10 gram/day food intake rate, if feeding on insects with total Hg concentrations such as those found in Clear Lake aquatic insects (0.012 – 0.5 ppm),

would be ingesting 5 to 20 times the mammalian Hg NOAEL of 0.16 mg/kg/day used in the GLWQI model. At the 29th North American Symposium on Bat Research last year, Massa and Grippo reported that insectivorous bats in areas under fish advisories had elevated tissue Hg levels, and that the body burden increased with age. Of the 40 bats sampled, 8 had fur or tissue Hg levels that exceed the USFWS hazard level (Massa and Grippo 1999).

Comment (Page 42): The endocrine-disrupting potential of MeHg in birds is so far unreported, however, work with fish and invertebrates suggest that some investigation is warranted, as does the skewed sex ratio of the egrets in the Spalding & Frederick study. Kernaghan and coworkers reported that freshwater mussels exposed to MeHg in the water column or via the diet exhibited significant bioaccumulation and decreased estradiol concentration following dietary exposure of 10 ng/L (Kernaghan et al. 1999). For a period of 6 months, two groups of walleye were reared on untainted catfish fillets, while two test groups were fed fillets injected with methylmercury, one group receiving 0.1 mug Hg g-1 food (low-mercury diet) and the other receiving 1.0 mug Hg g-1 food (high-mercury diet). After the exposure period, fish fed the low- and high-mercury diets had mean body burdens of 0.254 : 0.015 mug Hg g-1 and 2.37 : 0.09 mug Hg g-1, respectively. Dietary mercury significantly impaired both growth and gonadal development in males, which was apparent as reduced fish length, weight, and gonadosomatic index. Testicular atrophy was observed in fish fed the mercury-tainted fillets, but was nonexistent in control animals. Mercury also suppressed plasma cortisol in juveniles (sexes combined). Furthermore, these results suggest that methylmercury might also affect reproductive potential of teleosts by impairing testicular development in young (Friedmann et al. 1996). Some recent articles from the human health literature may provide helpful inputs for model development (Chapman 2000, Ginsberg 2000).

Response: Bats, as well as other Everglades species are important. However, funds do not permit investigation of many species. Research into the effects on wading birds, species of considerable importance to the public, is seriously underfunded. DEP has been seeking federal funds for work on endocrine disrupters for some time.

Comment (Page 43): Much of the Hg problem in the Everglades has focused on plant communities, fish, and birds, with relatively little attention devoted to other receptors. What are the levels of Hg in developing alligator embryos and young, and Ospreys and Eagles, and in raccoons and bats? Are alligators and raccoons eaten by subsistence peoples, as they are in other regions in the South, and should this be a consideration?

Response: The District is participating in a study with USFWS-Loxahatchee to collect young alligators from the northern to the southern Everglades and to quantify the THg and MeHg residues in their organs and tissues. These data will aid in quantifying exposure as a function of location, in developing a simplified bioaccumulation model, and in quantifying the dose-response relationships of individual organs using histopathological endpoints and accumulated concentrations

Comment (Page 43): The role of exotic and invasive plants in exacerbating the Hg problem by methylation needs to be considered. While not directly related to the Hg problem, the relationship between exotic and native fish as a food source for wading birds should be examined, as well as the relative levels of Hg in exotic and native fish of comparable size (i.e. the threat to consumers from prey fish).

Response: This is related to determining food web dynamics and exposure. Exotics will necessarily be considered as this work progresses.



Comment (Page 43): Quantitative measurements and estimates of fluxes except atmospheric deposition are few in the report. It is therefore strongly recommended that such data are presented in future reports.

Response: This is being done as part of the field work for development of the E-MCM.

Comment (Page 44): The stable mercury isotope *in situ* mesocosm experiments (being initiated by Gilmour and Krabbenhoft within the ACME project) are critical to developing a process-level understanding of the role that sulfur, phosphorus, and organic carbon play in mercury methylation and subsequent bioaccumulation. These experiment should be the “cornerstone” of continued work on mercury dynamics within the aquatic ecosystem of the Everglades. One way to accomplish the interactive relationship between process experimentation like this and the E-MCM development would be to use the E-MCM to model and thereby interpret the results of these mesocosm experiments.

Response: DEP and SFWMD concur. Every effort is being made to continue the close interaction between the field researchers and the modelers.

### **Panel Recommendations for Changes in this Report with References to Changes Made**

Comment (Page 5): Growers in the EAA should be informed of the role of sulfate-sulfur in the biogeochemical production of methyl mercury, and that they should begin to think of BMPs in the use of sulfur-bearing fertilizers and soil amendments.

Response: The DEP primary strategy is control of atmospheric emissions. Should this prove to be insufficient, management of production of methylmercury would be considered. However, evidence is lacking that abatement of sulfate from EAA agricultural practices would affect Everglades methylmercury bioaccumulation. Under these circumstances, it would not be appropriate to ask growers to consider sulfate BMPs.

Comment (Page 34): Within the appendices, however, some critical information was sometimes lacking regarding sample protocol, sample sizes, detection limits, and variance around means.

Response: Chapter 7 appendices have been reviewed to identify those to which this statement is applicable and appropriate text was added to provide the missing information if it was available.

Comment (Page 35): The clear trend in decreasing Hg levels in Largemouth Bass and egrets (as measured by feathers) is so striking that some very large source of MeHg must have decreased at the same rate a few years earlier, yet this is not evident from the data presented. It seems particularly unrelated to atmospheric deposition, unless there is a lag phase. This important relationship deserves more explanation in this section.

Response: A comment about the time lag between the reduction in source emissions in south Florida and the appearance of this trend has been added to the Chapter text at the end of the section entitled, RESPONSE OF NATURAL SYSTEM TO SOURCE REDUCTION. In addition, DEP offers the following response:

Peer reviewers and others have requested more information and a more fully developed analysis of the trend data included here. In a concurrent and somewhat overlapping effort, the Department is presently conducting an 'Atmospheric Mercury TMDL Pilot Study' that will satisfy this request, but it was not complete at the time of this writing. The TMDL Pilot study is a joint project between DEP; USEPA and its offices of Water, and Air and Radiation; and SFMSP scientists. Appendices 7-3 and 7-6 are the drafts for peer review of the water and air modeling support documents, respectively; these are being modified in response to comments. The final TMDL Pilot Study report will incorporate the revised modeling information, include additional trend information; and include modeling analyses of such data as exist regarding historical mercury emissions and deposition. We anticipate a draft final report of the TMDL Pilot Study in November, 2000, with subsequent publication by EPA following that agency's review and legal and policy analyses. Copies of the draft final report will be sent to all ECR Peer Reviewers and to interested parties. The 2002 Everglades Consolidated Report will contain further data on mercury trends, along with a summary of the TMDL Pilot Study final report.

Comment (Page 36): The report suggests that the relationship between sulfate levels and mercury bioavailability is complex, may not be linear, and may both stimulate and reduce bacterial methylation, depending upon levels. The report indicates that agricultural sulfur enhances MeHg production and bioaccumulation, but that sulfide inhibits Hg uptake by methylating bacteria. Thus, in the parts of the Everglades with the highest eutrophication, sulfide accumulation inhibits MeHg production. The balance between sulfate load and sulfide accumulation is thus a crucial factor in controlling MeHg production throughout the Everglades. Consistent with this, MeHg concentrations in all matrices were highest in central Everglades, but were lower in both the most pristine areas and in the most eutrophic areas (WCA2A, ENR). The exploration of the dynamics of the sulfur chemistry in the central Everglades is an important aspect of the on-going research.

Comment (Page 41): The factors that affect mercury methylation remains a key issue. Mercury methylation is very complex, and a greater understanding of the relationship between phosphorus and methylation is required. The data suggest that reducing P levels may increase Hg within fish and other biota within the Everglades. Adaptive management, in an iterative manner, will be required to track declines in P and changes in biodilution of Hg and biota.

Comment (Page 41): One of the key aspects in understanding Hg cycling within the Everglades presumably is the relationship between sulfur, phosphorus, and methylation by bacteria. This is critical to setting the phosphorus standard, but also in terms of agricultural interests, and its regulation. Therefore, further research on the relationship between phosphorus, sulfur and methylation by bacterial is absolutely critical to any consideration of the Hg problem and to food chain accumulation.

Response: In response to these remarks, new text has been added to the Section, MANAGEMENT IMPLICATIONS under Relationships to Other Water Quality Variables.

End – Chapter 7 Response to Comments.